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Original Paper

Reproducibility of Histological Diagnosis of Breast Lesions: Results of a Panel in Italy

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Sixteen pathologists independently examined and classified a series of 81 breast lesions selected from the files of several Italian Pathology Departments in the context of a national task force on breast cancer (FONCAM). A four category classification system was used for analysis; according to the majority diagnosis (MD), simply defined as the most frequently reported in the panel, the series included 37 benign lesions without atypia (45.7%), nine atypical hyperplasias (11.1%), 18 *in situ* (22.2%) and 17 invasive carcinomas (21.0%). Concordance, estimated for all possible pair-wise comparisons between pathologists, was good (mean kappa value: 0.59). A comparison between the diagnoses of each pathologist and the panel majority diagnosis was also made. Overall, a global kappa value of 0.72 was found (range 0.57–0.85), with category-specific values being excellent for invasive carcinoma (0.89) and benign lesions without atypia (0.77), relatively good for *in situ* carcinoma (0.69) but poor for atypical hyperplasia (0.38). These results confirm that quality assurance procedures are particularly indicated for large screening programmes for breast cancer, and suggest that for atypical lesions strict diagnostic criteria should be adopted.

Key words: breast lesions, atypical hyperplasia, breast cancer, reproducibility, histology, diagnostic accuracy

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INTRODUCTION

BREAST CANCER is the most common tumour and the leading cause of cancer death in the female population worldwide [1, 2]. A reproducible histological definition of different breast lesions, ranging between benign disease and invasive cancer, is necessary in order to treat individual patients correctly. Different histological classifications are commonly used for breast disease by groups of pathologists. Furthermore, diagnostic interobserver variability within the same classification is found [3].

In situ carcinomas are increasing because of more widespread early diagnosis and screening procedures [4]. In contrast, atypical hyperplasia (AH) is a rare condition (about 2–4% of all benign lesions) but because of the high risk of subsequent invasive breast cancer (BC) observed in several studies, it is considered an indication for a closer follow-up of patients [5–7]. The need of a common standardised terminology is one of the major issues in the field of breast pathology.

In recent years, the value of assessing the reproducibilty of diagnoses in pathology has become widely accepted [3, 8]. A procedure often used by pathologists in order to test their diagnostic reproducibility is to circulate a set of slides. Several reports have been published in the current literature, regarding quality assurance practices in diagnostic surgical pathology [9–20]. Traditionally, concordance between observers has been assessed by use of the kappa statistic [21], which includes a correction for the amount of agreement expected by chance alone. Recently, new approaches have also suggested the possibility of taking into consideration the individual characteristics of a specific observer [22].

The aim of this study was to evaluate the reproducibility of histological diagnoses in a series of 81 selected breast lesions among 16 pathologists in Italy.

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MATERIALS AND METHODS

Selection of histological material

Histological slides of breast lesions were selected in 16 local Pathology Departments in Italy. The selection was performed with the aim of including a representative sample of all the major categories of breast pathology. The participating members were from the University or Hospital Pathology Departments located in Arezzo (three pathologists), Bari, Bologna, Firenze, Genova, Leno, Mantova, Milano (three), Torino (three) and Trieste. Only some of the participants are primarily involved in diagnostic breast pathology. A set of 82 haematoxylin–eosin stained slides (usually one for each case) were randomly numbered and circulated among participants after an initial meeting in which general criteria were discussed. One specimen (no. 10) was damaged during the circulation and was not considered. Pathologists were asked to reach a diagnosis for each case and to report it in a form for the concordance analysis. No information about age, the original diagnosis or the diagnosis of the other pathologists was provided to the participants.

Classification

Pathologists reported their diagnoses for each case on a study form in which five diagnostic categories were available. The benign breast disease (BBD) classification originally proposed by Dupont and Page [23] and supported at the consensus meeting of the College of the American Pathologists [24] was modified, combining the three main categories of BBD into two: non-proliferative or proliferative lesions without atypia (A) and atypical hyperplasia (B). Two additional categories for *in situ* carcinoma were utilised [ductal non comedo and lobular (C) and ductal comedo (D)]. A fifth category was used for invasive carcinoma (E). Description of each specimen was allowed but only the final diagnostic decision (coded 1 to 5) was considered as follows:

A. Non-proliferative or proliferative without atypia benign breast disease;
B. Ductal and/or lobular atypical hyperplasia (AH);
C. Ductal non comedo and/or lobular *in situ* carcinoma;
D. Ductal comedo *in situ* carcinoma;
E. Invasive carcinoma.

The original forms were first checked for completeness and consistency and then computerised for the analysis. No correction was allowed for errors which might have occurred when pathologists reported their diagnoses (i.e. using wrong codes) once the original forms were obtained.

Analysis

Analysis was carried out using four categories, after combining all *in situ* carcinomas (1 = A; 2 = B; 3 = C+D; 4 = E).

The histological diagnoses of each pathologist were first compared to those of all the other 15. Overall, 120 such pairwise comparisons were performed ($n^*(n-1)/2$, where $n = 16$ readers). No attempt to reach a final consensus among panel members was made. The diagnoses of each pathologist were therefore compared to the MD, simply defined as the most frequent diagnosis expressed for each slide, and a category-specific analysis was also performed. As measure of agreement, either between the 16 pathologists and between each pathologist and the MD, the kappa statistic was used [21], which includes a correction for the amount of agreement which would be expected by chance alone. Values of kappa near the maximum of 1 indicate perfect agreement. Traditionally, values higher than 0.40 are considered acceptable and higher than 0.75 excellent. Standard errors (S.E.) of the overall kappa values were calculated according to Fleiss and associates [25].

The specimens were circulated only once; therefore no data were available on intra-observer reproducibility.

RESULTS

The distribution of the diagnoses for each of the 81 breast lesions and the resulting MD are shown in Table 1: 37 benign (45.7%), 9 AH (11.1%), 18 *in situ* carcinomas (22.2%) and 17 invasive carcinomas (21.0%). All 16 pathologists reached the same diagnostic conclusion in 22 cases (27.2%): 11 were classified as invasive carcinoma, one as *in situ* carcinoma and

Table 1. *Distribution of individual histological diagnoses* (16 *pathologists*) *and resulting majority diagnosis of* 81 *breast lesions* (*FONCAM, Italy*)

Case	Benign*	AH	*In situ*	Invasive	Majority diagnosis‡
1	3	8	4	1	2
2	0	9	7	0	2
3	0	0	0	16	4
4	0	0	0	16	4
5	0	1	13	2	3
6	12	1	3	0	1
7	16	0	0	0	1
8	0	0	0	16	4
9	0	0	0	16	4
10†	—	—	—	—	—
11	4	0	0	12	4
12	15	0	0	1	1
13	0	3	13	0	3
14	6	10	0	0	2
15	15	1	0	0	1
16	11	5	0	0	1
17	12	4	0	0	1
18	16	0	0	0	1
19	1	4	10	1	3
20	15	0	1	0	1
21	0	0	0	16	4
22	0	0	1	15	4
23	1	1	14	0	3
24	14	1	1	0	1
25	14	1	1	0	1
26	10	6	0	0	1
27	10	5	1	0	1
28	15	1	0	0	1
29	1	1	8	6	3
30	16	0	0	0	1
31	12	3	1	0	1
32	0	0	0	16	4
33	1	0	1	14	4
34	8	7	1	0	1
35	9	6	1	0	1
36	15	0	1	0	1
37	14	1	1	0	1
38	5	6	5	0	2
39	16	0	0	0	1
40	1	2	11	2	3
41	0	0	0	16	4

Table 1 continued overleaf

Table 1. Continued

Case	Benign*	AH	*In situ*	Invasive	Majority diagnosis‡
42	0	0	0	16	4
43	16	0	0	0	1
44	1	3	12	0	3
45	16	0	0	0	1
46	13	3	0	0	1
47	9	4	0	3	1
48	1	2	13	0	3
49	4	7	5	0	2
50	0	2	13	1	3
51	0	0	14	2	3
52	0	0	0	16	4
53	16	0	0	0	1
54	0	0	16	0	3
55	2	6	8	0	3
56	3	3	10	0	3
57	0	0	1	15	4
58	8	7	1	0	1
59	0	0	0	16	4
60	3	10	3	0	2
61	16	0	0	0	1
62	15	1	0	0	1
63	16	0	0	0	1
64	13	3	0	0	1
65	0	5	11	0	3
66	10	5	1	0	1
67	0	2	14	0	3
68	0	0	9	7	3
69	0	0	0	16	4
70	3	1	12	0	3
71	3	0	0	13	4
72	15	0	1	0	1
73	15	0	1	0	1
74	16	0	0	0	1
75	1	1	6	8	4
76	4	10	2	0	2
77	0	4	12	0	3
78	5	10	1	0	2
79	1	7	6	2	2
80	15	1	0	0	1
81	11	5	0	0	1
82	14	1	1	0	1

*Benign lesion without atypia. †Damaged during the panel. ‡1, benign lesion without atypia; 2, atypical hyperplasia; 3, *in situ* carcinoma; 4, invasive carcinoma. AH, atypical hyperplasia.

10 as benign disease without atypia, but in no case was AH diagnosed by all 16 participants. In the remaining 59 cases, at least one panellist was not in agreement with the others: all the four available diagnostic categories were used for 6 cases, three categories in 26 cases and two categories in the remaining 27 cases.

In the latter group, the same diagnostic conclusion was reached for 11 cases by 15 of the 16 pathologists, while only one was not in agreement (in 4 cases 15 panellists reported a diagnosis of "benign without atypia" versus 1 diagnosis of "AH"; in 4 cases 15 benign without atypia versus 1 *in situ* carcinoma; in 1 case 15 benign without atypia versus 1 invasive carcinoma; in 2 cases 15 invasive versus 1 *in situ* carcinoma).

Overall, only 9 cases were diagnosed as AH by the majority: in 2 such cases, all the four available diagnostic categories were reported, in 5 cases three categories and in 2 cases two categories.

Kappa values for all possible pair-wise comparisons between the 16 pathologists are shown in Table 2. The mean and median kappa values are also reported for each pathologist. In these 120 pair-wise comparisons, kappa values ranged between a minimum of 0.37 and a maximum of 0.91. Mean overall kappa values for each pathologist ranged between 0.48 and 0.66. The overall mean kappa value was 0.59.

In Table 3 the cumulative distribution of the diagnoses of all the participants versus the MD is reported. Overall, 1296 (16×81) diagnostic comparisons were performed: 1042 (80.4%) were in perfect agreement. Category specific crude agreement was 84% (499/592) for the benign lesion without atypia category, 53% (77/144) for AH, 74% (213/288) for *in situ* carcinoma and 93% (253/272) for invasive carcinoma. The global kappa value was 0.72. On nine occasions, a case was classified by an individual reader as benign lesion without atypia while the resulting MD was invasive carcinoma (cases no. 11, 33, 71 and 75 in Table 1 with four, one, three and one diagnoses, respectively). A careful review of the 2 cases with multiple benign diagnoses after the data analysis showed morphological patterns consistent with the diagnosis of tubular carcinoma (case 11) and ductal invasive carcinoma (case 71). In the latter, it was also noted that two sections of different areas of the surgical specimen were available for review on the same slide and only normal (non-malignant) tissue was present in one section. In contrast, four diagnoses of invasive carcinoma were followed by a MD of benign lesion without atypia (cases 12 and 47, with one and three diagnoses, respectively).

In Table 4, category-specific and global kappa values estimated for individual comparisons between each pathologist and the MD are reported; mean values for category specific and overall kappa statistics are also shown. Global kappa values for individual pathologists varied between 0.57 and 0.85. The category-specific kappa value was lowest for atypical hyperplasia (range 0.14–0.70; mean value 0.38) and highest for invasive carcinoma (range 0.78–1.00; mean value 0.89). Benign lesions without atypia and *in situ* carcinoma showed intermediate values (range 0.62–0.95 and mean value 0.77; range 0.40–0.89 and mean value 0.69, respectively).

DISCUSSION

Reproducibility of breast histological diagnoses was shown to be relatively good in this large panel. In particular, an excellent agreement was found for the diagnosis of invasive carcinoma and of benign lesions without atypia. However, in the two intermediate categories (AH and *in situ* carcinoma), serious difficulties emerged resulting in a very low kappa value for AH, while the diagnosis of *in situ* carcinoma was shown to be more consistent. In none of the 9 cases with a MD of AH was the same diagnostic conclusion simultaneously reached by all participating pathologists, in agreement with other previous reports [3]. These findings suggest that no clear definition of AH is currently available and pathologists give a different meaning to this diagnosis. Because of the low agreement obtained for this specific category, it has been recently suggested, but not widely accepted, that terminology such as "mammary intra-epithelial neoplasia" combining hyperplasia, AH and *in situ* carcinomas is adopted [3].

Not all diagnostic errors affect patient care to the same

Table 2. Concordance for all possible pair-wise comparisons between 16 pathologists; median, mean and overall mean kappa values for each pathologist (FONCAM, Italy)

	3	5	10	12	14	15	16	17	18	20	21	23	26	27	28	30	
5	0.56																
10	0.54	0.71															
12	0.48	0.68	0.64														
14	0.56	0.70	0.60	0.58													
15	0.47	0.62	0.59	0.48	0.50												
16	0.56	0.61	0.67	0.69	0.56	0.53											
17	0.63	0.71	0.67	0.73	0.61	0.48	0.68										
18	0.53	0.57	0.53	0.46	0.56	0.38	0.55	0.57									
20	0.61	0.71	0.68	0.67	0.68	0.58	0.64	0.70	0.52								
21	0.49	0.67	0.64	0.57	0.57	0.64	0.58	0.59	0.41	0.65							
23	0.57	0.62	0.48	0.53	0.60	0.54	0.57	0.66	0.50	0.59	0.46						
26	0.39	0.53	0.51	0.48	0.49	0.46	0.49	0.46	0.43	0.53	0.52	0.37					
27	0.56	0.69	0.61	0.62	0.71	0.55	0.62	0.71	0.44	0.79	0.61	0.65	0.52				
28	0.50	0.73	0.64	0.61	0.62	0.59	0.60	0.61	0.49	0.76	0.59	0.57	0.58	0.68			
30	0.60	0.76	0.66	0.72	0.63	0.51	0.67	0.91	0.49	0.68	0.60	0.70	0.42	0.73	0.63		
Median	0.56	0.67	0.64	0.58	0.60	0.53	0.60	0.66	0.50	0.67	0.59	0.57	0.49	0.62	0.61	0.66	Overall
Mean	0.54	0.66	0.61	0.60	0.60	0.53	0.60	0.65	0.49	0.65	0.57	0.56	0.48	0.63	0.61	0.65	0.59

Table 3. Cumulative distribution of histological diagnoses of 81 breast lesions in the comparison between majority diagnosis and all the 16 pathologists (FONCAM, Italy)

	Majority diagnosis				
All pathologists	Benign*	AH	*In situ*	Invasive carcinoma	Total
Benign*	499	31	14	9	553
AH	72	77	40	1	190
In situ	17	33	213	9	272
Invasive carcinoma	4	3	21	253	281
Total	592	144	288	272	1296

*Benign lesion without atypia. AH, atypical hyperplasia.

degree: minor disagreements may have minimal or no effect at all on patient care, while major discrepancies could seriously affect the patients. Either under-diagnosis or over-diagnosis may occur. In this study, four specimens were classified as benign lesions without atypia by a total of nine pathologists (four in 1 case, three in another) while the resulting MD was invasive carcinoma. The opposite was found in 2 different cases: a total of four pathologists (three in 1 case and one in the other) classified these specimens as invasive carcinoma while the panel MD was benign lesions without atypia. These major disagreements might have seriously affected the therapeutical decisions in individual patients.

All the slides were haematoxylin–eosin stained but were retrieved from the archives of 16 Pathology Departments in Italy and the technical quality of the different specimens might not have been optimal in every case. Other factors could have influenced the results. When the pathologists sampled the slides for this study, they might have included 'problem' cases more frequently than the routine daily diagnostic session of a general surgical pathologist. The artificial reading conditions might have also contributed to the disagreement, in particular because the series tended to be large and the time available for the classification was relatively short.

Table 4. Category-specific and global kappa values estimated for individual comparisons between each pathologist and the majority diagnosis of 81 breast lesions. Mean values for specific and overall kappa (FONCAM, Italy)

	Category-specific kappa values				
Pathologist	Benign*	AH	*In situ* carcinoma	Invasive carcinoma	Global kappa (S.E.)
3	0.62	0.37	0.63	0.89	0.63 (0.06)
5	0.95	0.65	0.72	0.93	0.84 (0.07)
10	0.75	0.25	0.82	0.92	0.75 (0.07)
12	0.85	0.29	0.69	0.92	0.76 (0.07)
14	0.72	0.41	0.84	0.89	0.72 (0.06)
15	0.73	0.37	0.52	0.78	0.64 (0.07)
16	0.85	0.14	0.64	0.89	0.71 (0.07)
17	0.80	0.25	0.77	0.96	0.75 (0.07)
18	0.67	0.27	0.45	0.82	0.58 (0.07)
20	0.87	0.70	0.89	0.88	0.85 (0.07)
21	0.80	0.54	0.67	0.85	0.74 (0.07)
23	0.64	0.27	0.64	0.82	0.62 (0.06)
26	0.63	0.24	0.40	0.88	0.57 (0.07)
27	0.75	0.44	0.86	0.88	0.75 (0.07)
28	0.87	0.47	0.72	1.00	0.80 (0.07)
30	0.80	0.31	0.73	0.92	0.73 (0.07)
Mean (S.E.)	0.77 (0.10)	0.38 (0.15)	0.69 (0.14)	0.89 (0.05)	0.72 (0.08)

*Benign lesion without atypia. AH, atypical hyperplasia.

In conclusion, we consider quality assurance in pathology mandatory, particularly in the context of a screening programme, when a higher proportion of *in situ* and more differentiated invasive cancers is expected. Our results suggest that agreement on the morphological characteristics of atypical lesions should be improved among pathologists in order to guarantee a homogeneous diagnostic classification and treatment to patients with a borderline breast lesion.

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APPENDIX

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