Papnet-assisted cytological diagnosis intensifies the already marked variability among cytological laboratories

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Slides were also provided by 20 Italian Laboratories of the Italian National Health Service: S. Filippo Neri Hospital (Rome), Gallipoli Hospital (Lecce), Health Authority n. 75 (Milan), Palmanova Hospital (Udine), Cytopathology Unit (Aosta), Infermi Hospital - Rimini (Forlì), Cytopathology Unit (Catania), Health Authority n. 28 - Vimercate (Milan), Health Authority n. 9 (Sondrio), Health Authority n. 10 - Camerino (Macerata), Health Authority n. 3 - Sestri (Genova), Health Authority n. 8 (Cagliari), Health Authority Castellammare (Naples), HealthAuthority (Ravenna), Health Authority Viareggio (Lucca), Infermi Hospital (Viterbo), Health Authority (Terni), Health Authority n. 8 (Foggia), Maggiore della Carità Hospital (Novara), Health Authority n. 11 (Venezia).

Summary

Objective: The main objective was to assess the sensitivity, specificity and reliability of PAPNET-assisted diagnosis in comparison with conventional screening.

Setting: Seven Italian and one English University or Research Institutes, and a random sample of an other 20 Italian Laboratories of the Italian National Health Service (INHS) provided the cervical smears.

Methods: During the training phase every center examined in rotation four sets of slides for a total of 300 representative slides. Afterwards, 900 "positive" slides were added to the 3,100 slides which were collected consecutively without any selection or exclusion. The eight main centers were divided into four couples and each couple of centers examined 775 slides with the PAPNET system, "blindly" to the original diagnosis. An expert cytopathologist (M.A.) of the National Institute of Health (NIH) reassessed 40% of the slides with an original negative diagnosis to evaluate the false negative rate. Two expert NIH cytopathologists (M.A., G.M.) re-examined all slides where a disagreement had been observed between the original and one or both of the study diagnoses. The main analyses concerned the following three main categories: WNL and unsatisfactory for evaluation; ASCUS, AGUS and LSIL; HSIL and carcinoma. A special algorithm was devised to define the reference diagnosis for sensitivity and specificity assessment.

Results: Laboratories, even belonging to the same couple, classified as "no review" a very different proportion of slides ranging from 35% to 74%. The index of kappa agreement between the members of couples examining the same sets of slides was low or very low, ranging from 0.30 to 0.03. The sensitivity of the review classification was particularly low in some laboratories. Surprisingly, only a small correlation was observed between the sensitivity of the review classification and the proportion of slides classified as "review".

The "tentative" diagnosis on PAPNET tiles of the "review" slides was almost as reliable as the microscopic diagnosis.

In the overall performance, there were many significant differences among the eight laboratories. The best laboratory had a sensitivity of 95% and a specificity of 96%. At least three laboratories displayed unacceptably low sensitivity and one a very low specificity.

Conclusion: Altogether these results seem to confirm that there are wide differences among cytological laboratories per se, and that these differences are intensified by the use of an instrument like PAPNET. The huge variation in performance may be explained by differences in basic skills and by different training, but it is difficult to understand exactly what could have been done to reduce it.

Key words: Pap-net; Screening; Cervical Cytology; Inter-observer variation; Diagnosis validation.

Introduction

Well-organised cervical screening programmes have been shown to be effective in reducing both incidence and mortality of invasive cervical carcinoma [1-3].

Cervical screening smear examination is intensive, repetitive work requiring continuous and demanding concentration by trained staff and quality control procedures [4-8].

In cervical screening, the risk of false negative reports has been cause for concern. The false negative rate has been reported to vary between 5 and 55% and this variation is only partially explained by the different methods which were used to measure the sensitivity [6, 7, 9-12].

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Among the measures to reduce the risk of false negatives, a 10% random re-screening has been widely adopted in cytology laboratories in the United States for many years in order to comply with the Clinical Laboratory Improvement Acts (CLIA) of 1988.

This technique has been notably criticised by some authors [13, 14] as an inefficient system of quality control as only a fraction (10%) of total negative smears processed in a laboratory are re-screened.

The other system is a rapid review of negative smears under a low power microscope (x10) and is currently adopted in the U.K. The equipment that is the object of this paper has been introduced mainly as a help to reduce false negatives.

Objectives and main features of the project

The main objectives of this research project were the following: 1) to assess the sensitivity, specificity and reliability of the PAPNET-assisted diagnosis; 2) to assess the possibility of making a full diagnosis on the PAPNET tiles without microscopic confirmation; 3) to compare the variability of the microscopic and PAPNET-assisted diagnosis.

Material and Methods

PAPNET system

PAPNET is a semiautomated and interactive testing system [15-17]. It consists of two major subsystems: *the scanner* or screening apparatus and *the review station* [18-22].

The examiner is asked to divide the slides into two categories: "Review" and "No review". No review slides are reported as negative and review slides are checked with an optical microscope.

The potential advantages of the system as a complement to traditional screening can be summarised as follows:

- Abnormal cells are selected by the program from different parts of a smear;
- The cytoscreener views static images, thus reducing the likelihood of fatigue-related errors.
- Less time is needed to screen the same number of slides.

Main features of the present study

The present study has the following main features:

– It is multicentric and includes two different kinds of laboratories: a group of eight "main" laboratories, all belonging to University or Research Institutes, one in London, UK and seven in Italy: Turin, Trieste, Pietra Ligure, Pisa, Rome (two Centres), Naples and a probabilistic random sample of 20 smaller NHS laboratories (6,000-10,000 cervical smears examined per year). All University and Research Institute laboratories had their own PAPNET "review station" The contribution of the other labs was limited to the provision of slides.

– Examiners were doubly blinded (to the original diagnosis and to clinical information);

– Almost 50% of "review" slides were re-examined by two cytopathologists.

Slide selection. Each University-based laboratory provided 200 slides, 150 consecutive slides from those examined in the previous few months and 50 positive; the Italian National Health Service laboratories only 75, 50 consecutive slides and 25 positive. The consecutive slides were taken without any selection or exclusion. The positive slides had to include all diagnostic categories (HPV, ASCUS, LSIL, HSIL, squamous-carcinoma, AGUS, adenocarcinoma). A total of 3,100 slides were collected.

For each slide the original cytological diagnosis was reported on a standardised form.

Of the 3,100 slides 26 slides with an original diagnosis of "inadequacy" were excluded from further processing.

Cases were identified through three bar-code labels: one placed on the slide, one on the form to be sent to the National Institute of Health and one on a log book. The slides were packed in special boxes containing 100 slides each and mailed to Amsterdam where the PAPNET central processing unit was located.

Tapes were prepared by the central unit and returned to the main laboratories together with the relevant slides according to the following protocols: the main laboratories were divided at random into four couples and each member of the couples was sent the images of the same 775 slides, their own 400 plus 375 which were provided by five of the 20 other laboratories.

Standardised form. An "ad hoc" form was designed to record the results. On the back of the form there was a list of diagnostic categories according to the Bethesda System with their code.

Training phase. Every University based laboratory (except for the already experienced Department of Cytopathology, St. Mary's Hospital Medical School, London) examined in rotation another four sets of slides (2 sets of 100 and 2 sets of 50 slides), for a total of 300 representative slides.

The 300 slides of this phase were the same for the seven Italian laboratories and were provided by the Cytopathology I, Department of Experimental Medicine, "La Sapienza" University of Rome.

PAPNET review station use. Every main laboratory examined the 775 slides "blindly" to the original diagnosis. Each slide was classified either as "no-review" or as "review". A tentative diagnosis on PAPNET tiles was requested for "review" slides before the final microscopic examination.

Control of negative slides. Forty percent of the slides with a negative original diagnosis were reassessed by an expert NIH cytopathologist (M.A.). To check the accuracy of this second examination 20 positive slides were randomly inserted in the set blindly to the examiner who had no possibility of identifying them. In all, 1004 slides were so reviewed.

Control of positive slides. Two expert cytologists of the NIH (M.A., G.M.) re-examined in double all slides where a disagreement between the original and one or two of the study diagnoses had been observed, also if the disagreement concerned only the degree of lesion severity. In total, 318 slides were so reviewed.

Regrouping of diagnoses. In order to compare cytological results between the PAPNET tentative diagnosis and conventional microscope reading the diagnostic categories were grouped into three principal categories: 1 = Normal (Unsatisfactory for evaluation + Within normal limits and benign cellular changes); 2 = ASCUS and AGUS + LSIL; 3 = HSIL + Carcinoma.

Also a six-category classification was used: 1 = Unsatisfactory for evaluation; 2 = Within normal limits and benign cellular changes; 3 = ASCUS and AGUS; 4 = LSIL; 5 = HSIL; 6 = Carcinoma.

Interlaboratory and intralaboratory agreement.

To assess the within and between laboratory agreement, we used the kappa statistic. Weighted kappa, which takes into account the magnitude of the differences, was substituted for simple kappa when the categories were more than two [23].

Reliability was assessed between members of couples of laboratories for the classification "review" - "no review", and for the microscopic diagnosis, restricting the comparison to the slides which had been judged "review" by both.

Accuracy estimation. The "reference" or "gold diagnosis" was so defined:

– *Negative original slides:* Original diagnosis; this decision was taken based on a check of the original negative slides (see results).

– *Positive original slides which were examined by microscope by three laboratories* (original one plus two): Agreed diagnosis if the diagnosis was the same in all three. NIH diagnosis if concordant with at least one of the other three diagnoses; the diagnosis made by two laboratories if INIH was discordant with everyone. If all four diagnoses were different, the slide was excluded.

– *Positive original slides which were seen by microscope by only two laboratories* (original plus 1): Agreement of diagnosis if both diagnoses were the same; NIH diagnosis if concor-

dant with at least one other lab. If all three diagnoses were different, the slides were excluded.

– *Positive original slides which were seen by microscope by only the original lab* (because both of the PAPNET laboratories assessed them as "non review"). The NIH diagnosis was considered as a reference.

PAPNET performance was also assessed calculating the proportion of "no review" slides which had a positive reference diagnosis (a kind of lack of sensitivity) and the proportion of review slides which had a negative reference diagnosis (a kind of lack of specificity).

Statistical analysis. Data were recorded using EpiInfo6 and an extensive process of data editing was performed [24].

Results

Overview of the results

After excluding 23 slides because they were deteriorated and too difficult to examine, 3,077 slides were processed; 2,541 (82.6%) with a negative and 518 (16.8%) with a positive original diagnosis. In less than 1% of the original diagnoses the diagnosis was "inadequate smear" (Table 1).

Six thousand one hundred and forty-four readings at PAPNET were performed, which produced 3,451 (56.4%) "no review" screening PAPNET diagnoses, 3,222 (63.4%) from original negative slides and 229 (26.6%) from original positive slides. The difference in proportion of the "no review" PAPNET screening diagnosis between negative and positive original diagnoses was of course highly significant.

Table 1. — *Distribution of the original 28 laboratory diagnoses (regrouped) after exclusion of 23 slides*

Diagnosis	Freq	%
Unsatisfactory for evaluation	18	0.6
Negative	2541	82.6
Positive	518	16.8
ASCUS and AGUS	91	
LSIL	220	
HSIL	174	
Invasive Carcinomas	33	
Total	3077	100.0

Control of negative slides

The cytopathologist was able to identify all the 20 "seeded" positive slides, which guarantees that also other positive slides incorrectly classified by the original lab as negative would have been detected. All original negative slides were confirmed as such, thus giving an agreement of 100%. We found a surprising specificity of 100%.

Agreement between laboratories which in the classification of slides in "review" and "no review" (so-called PAPNET triage)

Laboratories, even laboratories which received the same slides (1 and 2; 3 and 5; 4 and 8; 6 and 7) classified as "non review" found a very different proportion of slides - from 35% to 74%. The greatest difference (37%) was observed between labs 4 and 8 (Table 2).

Table 2. — *Distribution of "review" and "no review" classification in couples of laboratories which examined the same slides. Also the percentage of "no review" of all slides and of slides with an original negative diagnosis are reported. All differences between couples of laboratories are highly significant ($p < 0.0001$).*

Laboratories number	N of slides	No review	Review	% of no review on all slides	% of no review on original neg. slides
01	764	532	232	69.6	82.1
02	764	350	414	45.8	54.0
03	766	566	200	73.9	89.8
05	766	417	349	53.7	74.7
04	772	290	482	37.6	48.7
08	772	578	194	74.9	97.1
06	764	268	486	35.1	40.5
07	764	454	310	59.4	68.6

The same order of differences was found if only the slides with an original "negative" diagnosis were considered (Table 2, last column).

Consequently, the index of kappa agreement between the members of couples receiving the same slides was low or very low, from 0.30 to 0.03 (Table 2). The kappa values were much better for slides with an original "positive" diagnosis (Table 3, last column), however even in his case one couple showed an agreement lower than 0.60.

Table 3. — *Index of raw agreement and kappa in the classification of "review" and "no review between couples of laboratories that examined the same slides (see Table 2 for numbers): the last column refers only to slides which were originally classified as positive.*

Laboratories	Index of agreement	Kappa	
		All slides	Only positive
01-02	0.63	0.28	0.81 (116 slides)
03-05	0.66	0.30	0.61 (136 slides)
04-08	0.46	0.03	0.55 (177 slides)
06-07	0.62	0.28	0.85 (102 slides)

Agreement between tentative and microscopic diagnosis

This section deals with the comparison between the "tentative" diagnoses with PAPNET, i.e. the diagnoses which were made by directly examining the tiles of the "review" slides.

There was a moderate variation among laboratories, with one couple having a weighted kappa as low as 0.55 (Table 4). The weighted kappa was never higher than 0.75.

Table 4. — *Reliability between couples of laboratories with PAPNET tentative diagnosis-only slides which were classified as "review" by both laboratories.*

Laboratories	No of slides	Index of agreement	Unweighted kappa	Weighted kappa
01-02	176	0.70	0.54	0.71
03-05	146	0.72	0.51	0.73
04-08	129	0.56	0.31	0.55
06-07	259	0.80	0.59	0.74

Table 5. — *Reliability between couples of laboratories in the final microscopic diagnosis - only slides which were classified as "review" by both laboratories*

Laboratories	*No of slides*	*Index ommf agreement*	*Unweighted kappa*	*Weighted kappa*
01-02	176	0.78	0.66	0.81
03-05	146	0.72	0.51	0.70
04-08	129	0.66	0.47	0.66
06-07	259	0.82	0.64	0.78

The reliability within couples improved with microscopic examination, except for one couple (Table 5).

Sensitivity and specificity of the eight main laboratories towards the reference diagnosis are summarised in Table 6a for the PAPNET classification "review - no review", Table 6b for the microscopic diagnosis of the "review slides" and Table 6c for the microscopic diagnosis of all slides.

Table 6. — *Specificity and sensitivity of the 8 main laboratories*

6a. *Review (all considered positive)/No review (all considered negative) intermediate PAPNET classification towards reference positive or negative diagnosis*

Laboratories	% of "review" slides	Sensitivity	95% confidence limits	Specificity	95% confidence limits
1	15.6	76.6	68.0-83.5	96.5	94.7-97.7
2	25.6	87.0	83.8-89.6	88.3	80.5-93.4
3	8.4	40.2	31.5-49.4	97.9	96.3-98.8
5	14.4	72.5	63.5-80.1	96.8	95.0-98.0
4	25.5	57.4	49.4-65.1	84.3	80.8-87.2
8	13.5	47.6	39.8-55.5	97.0	95.0-98.2
6	12.5	77.2	67.0-85.0	97.5	95.8-98.5
7	13.8	96.6	94.6-97.8	80.2	70.3-87.6

6b. *Microscopic results towards reference diagnosis in "review slides" only*

Laboratories	% of "review" slides	Sensitivity	95% confidence limits	Specificity	95% confidence limits
1	15.6	94.9	88.1-98.1	95.6	95.6-98.4
2	25.6	96.4	90.5-98.8	82.9	75.1-88.8
3	8.4	77.4	64.7-86.7	89.6	81.8-94.5
5	14.4	79.2	70.1-86.3	96.8	93.2-98.6
4	25.5	67.4	58.8-75.0	83.2	78.4-87.1
8	13.5	74.7	64.3-83.0	86.7	68.4-95.6
6	12.5	91.5	82.7-96.2	98.2	95.8-99.2
7	13.8	91.9	83.4-96.4	87.7	81.6-92.1

6c. *Microscopic results towards reference diagnosis in all slides*

Laboratories	Proportion of "review" slides	Sensitivity	95% confidence limits	Specificity	95% confidence limits
1	15.6	82.2	73.9-88.4	99.2	98.0-99.7
2	25.6	94.8	88.5-97.9	96.3	94.4-97.6
3	8.4	40.3	31.6-49.7	98.2	96.7-99.0
5	14.4	72.5	63.5-80.1	96.8	95.0-98.0
4	25.5	55.0	47.2-62.6	90.9	88.1-93.1
8	13.5	41.6	34.1-49.5	99.2	98.0-99.8
6	12.5	91.5	82.7-96.2	98.2	95.8-99.2
7	13.8	96.5	94.7-97.8	80.6	71.1-87.6

The sensitivity of the classification "review/no review" was relatively low (Table 6a), as low as 40% in lab 3. Only one lab, number 2, had really satisfactory results, with a sensitivity of about 97%, even if it was the one with the highest proportion of slides classified as "review". There was of course a correlation between the sensitivity and the proportion of "review" slides (first column of Table 6a), however it was low: Sperman Rho 0.38, p = 0.3.

Table 6b allows a comparison of the microscopic diagnoses of slides classified as "review". The results are in general good, however there was a certain amount of variation with sensitivity ranging from 96% to 67% and specificity from 98% to 83%. The low values of sensitivity were somewhat unexpected because of the greater attention that should have been paid to the "review" slides.

Overall performance is displayed in Table 6c. There were many significant differences among the eight laboratories as shown by the lack of overlapping of confidence intervals. The best laboratory was number 2, with a sensitivity of 95% and a specificity of 96%. At least three laboratories had unacceptably low sensitivities and one a very low specificity. Almost the same correlation as before was seen here between sensitivity and the proportion of review slides.

Discussion

In our study the great variation in the number of slides which were classified as "no review" and so practically as negative, and the relatively low sensitivity of the review classification toward the reference diagnosis are very unsatisfactory results for the PAPNET system.

Mitchell *et al.* [25] reported a PAPNET sensitivity of only 44% in the "review" classification of a set of slides with about 1% of positives cases (which had been confirmed histologically). At a second reading of the same slides the sensitivity increased to 84%, confirming the need and the efficacy of screeners' training.

Coleman *et al.* [26] found a similar sensitivity for PAPNET-assisted (82%) and conventional (83%) screening. Specificity was significantly better for PAPNET than conventional screening: 77% vs 42%. A huge variation among the five participating laboratories was observed.

Earlier Slagel *et al.* [5] reported in a single lab a much better sensitivity of PAPNET-assisted vs conventional screening (97% vs 75%), but with a lower specificity (81% vs 99%, respectively).

Altogether these results seem to confirm that there are wide differences among cytological laboratories per se, and that these problems are intensified by the use of new instruments like PAPNET. The huge variation in performance may be explained by differences in basic skills and by different training, but it is difficult to understand exactly what kind of training or what other actions could have been implemented to reduce the variation.

This study appears after the PAPNET system has been withdrawn from the market. It is a pity that it was not concluded before, because it might have either contributed to a earlier dismissal of the instrument or it might have prompted the makers to improve the system or to recommend more systematic training of the users.

We hope that this study may at least contribute to the methodology of the technological assessment of diagnostic instruments.

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References

[1] IARC Working Group on Cervical Cancer Screening and UICC Project Group on the Evaluation of Screening Programmes for Cancer of the uterine cervix: (eds.) Hakama M., Miller A. B., Dayne, IARC Scientific Publications n. 76, 1986.

[2] Franco E., Monsonego J. (eds).: "New Developments in Cervical Cancer Screening and Prevention". Oxford, Blackwell Science, 1997.

[3] Hakama M. In: "New Developments in Cervical Cancer Screening and Prevention". Blackwell Science, Oxford, 1997, 190.

[4] Koss L. G.: "The Papanicolaou test for cervical cancer detection. A triumph and a tragedy". *JAMA*, 1989, *269*, 737.

[5] Slagel D. D., Zaleski S., Cohen M. B.: "Efficacy of automated cervical cytology screening". *Diag. Cytopathol.*, 1994, *13*, 26.

[6] van Der Graaf Y., Voijs G. P.: "False negative rate in cervical cytology". *J. Clin. Pathol.*, 1989, *40*, 438.

[7] Mody D. R., Davey D. D., Branca M.: "Quality assurance and risk reduction guidelines". *Acta Cytol.*, 2000, *44*, 496.

[8] Wied G. L., Bartels P. H., Bibbo M. *et al.* Editorial: "Computer assisted quality assurance". *Acta Cytol.*, 1996, *40*, 1.

[9] Richart R. M.: "Evaluation of true false negative rate in cytology". *Am. J. Obst. Gynaec.*, 1964, *89*, 723.

[10] Rilke F., Mango L., Alasio L.: "Sensitivity of the PAPNET cytological screening system to a routine mixture of conventional smears". *Mod. Pathol.*, 1993, *6*, 31.

[11] Rombach J. J., Cranendonk R., Velthius F. J.: "Monitoring laboratory performance by statistical analysis of concordance". *Acta Cytol.*, 1987, *31*, 887.

[12] Ronco G., Montanari G., Aimone V., Parisio F. *et al.*: "Estimating the sensitivity of cervical cytology: errors of interpretation and test limitations". *Cytopathology*, 1996, *7*, 151.

[13] Jordan S. W.: "Great expectations: cytology provision of CLIA-88 and the role of professional societies". *Cytopathology Annual*, 1992, 235.

[14] Kaminsky F. C., Burke R. J., Haberle K. R., Mullins M. S.: "Rescreening policies in cervical cytology and their effect on detecting the truly positive patients". *Acta Cytol.*, 1995, *39*, 239.

[15] Ashfaq R., Salinger F., Solares B. *et al.*: "Evaluation of the PAPNET System for prescreening triage of cervico-vaginal smears". *Acta Cytol.*, 1997, *41 (4)*, 1058.

[16] Attwood M. E., Woodman C. B. J., Luesley D., Jordan J. A.: "Previous cytology in patients with invasive carcinoma of the cervix". *Acta Cytol.*, 1985, *29*, 108.

[17] Farsworth A., Chambers F. A., Goldschmidt C. S.: "Evaluation of the PAPNET System in a general pathology service". *M. J. A.*, 1996, *165*, 429.

[18] Kharazi M., Keyhani-Rofagha S., O'Toole R.: "Automated screening of cervical cytology using PAPNET: comparative cytological findings with biopsy". *Am. J. Clin. Pathol.*, 1993, *99*, 337.

[19] Kish J. K., Shelk D.: "Evaluation of the performance of the PAPNET automated cytological screening system on the conventionally prepared cervical Papanicolaou smears at Hinsdale Hospital". Hisdale, Illinois., *Anal. Quant. Cytol. Histol.*, 1992, *14*, 267.

[20] Klinkhamer P. J. J. M., Voijs G. P., de Haan A. F. J.: "Intraobserver and interobserver variability in the quality assessment of the cervical smears". *Acta Cytol.*, 1989, *33*, 215.

[21] Kok M. R., Boon M. S., Boon M. E.: "Effects of applying neural networks in cervical screening: lower overtreatment rates and less over-diagnosis for patients with mild/moderate dysplastic smears". *J. Cell. Pathol.*, 1996, *1*, 109.

[22] Koss L. G., Eunice Lin M. D., Schreiber K., Elgert P., Mango L.: "Evaluation of the PAPNET cytologic screening system for quality control of cervical smears". *Am. J. Cl. Path.*, 1994, *101*, 220.

[23] Landis J. R., Koch G. G.: "The measurement of observer agreement for categorical data". *Biometrics*, 1977, 33, 159.

[24] Fleiss J. L., Cohen J.: "Statistical Methods for Rates and Proportions". New York: John Wiley and sons, 1981.

[25] Mitchell H., Medley G.: "Detection of laboratory false negative smears by the PAPNET cytologic screening system". *Acta Cytol.*, 1998, *42*, 265.

[26] PRISMATIC Project Management Team. Assessment of automated primary screening on PAPNET of cervical smears in the PRISMATIC trial., *Lancet.*, 1999, *353*, 1381.

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