Triage of women with equivocal or low-grade cervical cytology results: a meta-analysis of the HPV test positivity rate

Marc Arbyn [a, b, c, *], Pierre Martin-Hirsch [d], Frank Buntinx [e], Marc Van Ranst [f], Evangelos Paraskevaidis [g], Joakim Dillner [c, h]

[a] *Unit of Cancer Epidemiology, Scientific Institute of Public Health, Brussels, Belgium*
[b] *ECCG (European Cooperation on development and implementation of Cancer screening and prevention Guidelines, IARC, Lyon, France*
[c] *CCPRB (Cancer Control using Population-based cancer Registries and Biobanking) Network, Lund University, Malmö, Sweden*
[d] *Central Lancashire Teaching Hospitals, Preston, UK*
[e] *Belgian Centre of Evidence-Based Medicine, Belgian Branch of the Cochrane Collaboration, University of Leuven, Belgium; Department of General Practice, University of Leuven, Belgium & University of Maastricht, The Netherlands*
[f] *Department of Clinical and Epidemiological Virology, University of Leuven, Belgium*
[g] *Department of Obstetrics and Gynaecology, University Hospital of Ioannina, Greece*
[h] *Department of Medical Microbiology, Lund University, Malmö University Hospital, Malmö, Sweden*

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Abstract

Consistent evidence underlines the utility of human papillomavirus (HPV) DNA testing in the management of women with equivocal cervical cytological abnormalities, but not in case of low-grade lesions. We performed a meta-analysis including studies where the high-risk probe of the Hybrid Capture-II is used to triage these two cytological categories. The triage test-positivity rate reflects the colposcopy referral workload.Data were pooled on the HPV test positivity rate in women with atypical squamous cells of undetermined significance (ASCUS/ASC-US) or low-grade squamous intraepithelial lesions (LSIL), derived from different cytological classification systems. The meta-analysis was restricted to studies, published between 1991 and 2007. A random-effect model was applied for meta-analytical pooling and the influence of covariates on the HPV positivity rate was analyzed by meta-regression. The variation by age was assessed within individual studies since age strata were not defined uniformly. On an average, 43% (95% CI: 40–46%) of women with ASCUS/ASC-US were high-risk HPV positive (range 23–74%). In women with LSIL, the pooled positivity rate was 76% (95% CI: 71–81%; range 55–89%). In spite of considerable inter-study heterogeneity, the difference in HPV positivity between the two triage groups was large and highly significant: 32% (95% CI: 27–38%). HPV rates dropped tremendously as age and cutoffs of test positivity increased. Other factors (cytological classification system, country, continent, collection method and year of publication) had no statistically significant impact, except in LSIL triage where HPV positivity was significantly lower in European compared to American studies. Women with LSIL, especially younger women, have high HPV positivity rates suggesting limited utility of reflex HPV triaging these cases. Research is needed to identify more specific methods to triage women with low-grade squamous cervical lesions.

Keywords: cervical cancer • atypical squamous cells of undetermined significance • low-grade squamous intraepithelial lesions • triage • human papillomavirus • HPV • meta-analysis

Introduction

Women with minor cytological lesions identified in a Pap smear have a small but significantly increased risk of developing cervical cancer compared to women with normal smears. Reviews of the natural history of cervical epithelial atypia or low-grade squamous lesions suggest that the 2-year cumulative risk of invasive cervical cancer is in the range of 0.10% to

*Correspondence to: M. ARBYN,
Unit of Cancer Epidemiology, Scientific Institute of Public Health,
J. Wytsmanstreet 14, B1050 Brussels, Belgium.
Tel.: 0032 (0)2 6425021
Fax: 0032 (0)2 6425410
E-mail: m.arbyn@iph.fgov.be

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0.25% [1, 2]. Therefore, careful follow-up of these lesions is warranted.

The recognition of the strong association between persistent infection with oncogenic human papillomavirus types and the subsequent development of cervical cancer has prompted the detection of HPV DNA as an alternative triage method [3]. A recent meta-analysis of the accuracy of HPV DNA detection for cervical intra-epithelial neoplasia of grade 2 or worse ($CIN2^+$), using the high-risk probe of the Hybrid Capture II assay (HC2, Qiagen, Gaithersburg, MD, USA), in case of equivocal cytology demonstrated that the pooled estimate of the sensitivity and specificity was 94.8% (95% CI [CI]: 92.7–96.9%) and 67.3% (95% CI: 58.2–76.4%), respectively [4]. The pooled sensitivity for predicting presence of $CIN3^+$ was 96.4% (95% CI: 93.5–99.9%) and the pooled specificity was 56.5% (95% CI: 45.5–67.5%) [5]. The sensitivity of the HC2 assay was 16% (ratio = 1.16; CI: 1.04–1.29%) and 13% (ratio = 1.13; CI: 1.05–1.22%) higher than that of repeat cytology at cut-off atypical squamous cells of unspecified significance (ASCUS)/borderline dyskaryosis or worse for, respectively, $CIN2^+$ or $CIN3^+$. The specificity of cytological and virological triage was similar.

Until recently, the recommended policy in case of cervical equivocal (borderline) or mild (low-grade) cytological abnormalities consisted of the repetition of the smear and referral for colposcopy if the lesion persists or progresses [6, 7]. Meanwhile, the ASCUS-LSIL triage study (AITS) has demonstrated superior performance of triage of women with ASCUS by testing for high-risk HPV types compared to repetition of the Pap smear or immediate colposcopy referral. The results from the ASCUS and LSIL triage study (ALTS), corroborated by meta-analytical work, provide the evidence for current recommendations for reflex hrHPV testing in case of atypical squamous cells of undetermined significance [8–11]. Recommendations for managing low-grade cytological abnormalities are not uniform, ranging from immediate colposcopy [10] to repeat cytology and referral if cytological abnormality is persistent [12]. The utility of reflex hrHPV testing in case of LSIL or mild dyskaryosis is more controversial: it is not recommended in the most recent guidelines of the American Society for Colposocopy and Cervical Pathology [10], whereas others consider introducing it in screening programmes [13]. In Australia, reflex HPV testing to triage minor cytological abnormalities was not yet accepted, but this recommendation has been criticized [14]. In this new systematic review, we compare the HPV test positivity rate in women with equivocal and low-grade cytological abnormalities and examine how triage can be optimized by targeting different age groups.

Methods

Methods for retrieving published reports regarding the accuracy of HPV triage of women with minor cervical abnormalities were described previously [4, 15]. Studies were included if the following three criteria were fulfilled: (1) women had an index smear showing ASCUS or low-grade intra-epithelial lesions (LSIL) and study outcomes were reported separately for both triage groups, (2) the high-risk probe of the Hybrid Capture II was applied using the standard cut-off as positivity criterion (signal, expressed as relative light units [RLU] more intense than that of a control sample which contains 1 pg of HPV DNA per millilitre) and (3) all women were submitted to verification with colposcopy and colposcopy-directed biopsies and/or endocervical curettage when presence of squamous or glandualar intra-epithelial neoplasia was suspected. From the ALTS [16, 17], we used results from two of the three trial arms: women randomly assigned to immediate colposcopy and women randomly assigned to the HPV DNA testing arm, where colposcopic verification was restricted to women being HPV positive or having HSIL cytology.

In the current meta-analysis, we focus on the proportion of women with a positive HC2 test in atypical and low-grade squamous lesions. For this reason, inclusion criteria were relaxed and studies on management of minor squamous cervical lesions, distinguishing atypia and low-grade abnormalities with partial gold standard verification and/or age-stratification of the HPV status were also included.

The 1991 and 2001 versions of The Bethesda System (TBS) and the Terminology of the British Society of Clinical Cytology (BSCC) were used for classification of cytology [18–20]. In the 1991 version of TBS, ASCUS encompassed three subcategories of atypical squamous cells: (a) favour reactive (ASC-R); (b) undetermined significance and (c) neoplasia cannot be excluded. In TBS-2001, the first subcategory (ASC-R) was lumped with 'negative for neoplasia or malignancy', whereas the second and third subcategories were identified as ASC-US (with hyphen; atypical squamous cells of undetermined significance) and ASC-H (atypical squamous cells, high-grade lesion cannot be excluded), respectively. For the current meta-analysis, we computed the number of ASCUS cases if possible from the respective subcategories. In publications, using TBS-2001, where this was not possible, only data on ASC-US cases were extracted. Studies reporting data exclusively on ASC-H or atypical glandular cells were excluded. The definition of low-grade squamous intraepithelial lesions (LSIL) remained unchanged in TBS-1991 and TBS-2001. The BSCC terms borderline cytology and mild dyskaryosis were considered as similar to ASCUS/AS-CUS and LSIL, respectively, but were treated separately [21].

We used a random effect model for pooling proportions [22]. Inter-study heterogeneity was assessed with Cochrane's Q-test [23]. The percentage of total variation across studies due to heterogeneity was evaluated by the I^2 measure [24]. Forest plots were drawn showing the variation of the HC2 test positivity rate among all studies together with the pooled measure [25, 26]. Subgroup meta-analyses were used to distinguish the respective cytological classification systems [27]. A random-effect was also used to pool the difference in test positivity rate between ASCUS/borderline and LSIL/mild dyskaryosis cases [28].

The change in HPV positivity rate by age category was assessed by a chi-square trend, which generalizes the Wilcoxon test to several ordered groups [29]. The influence of study characteristics on inter-study heterogeneity was explored using a multi-variate hierarchical meta-regression with the logit-transformed HPV positivity rate as the dependent variable [30–32]. Relative risks were computed from the coefficients of the meta-regression using established formulas [33, 34].

We used Stata, version 10.0 (Stata Corp., College Station, TX, USA), for statistical analysis [27, 35].

Results

We identified 32 studies enrolling all together 26,311 women with a cytological report of ASCUS, ASC-US or borderline dyskaryosis

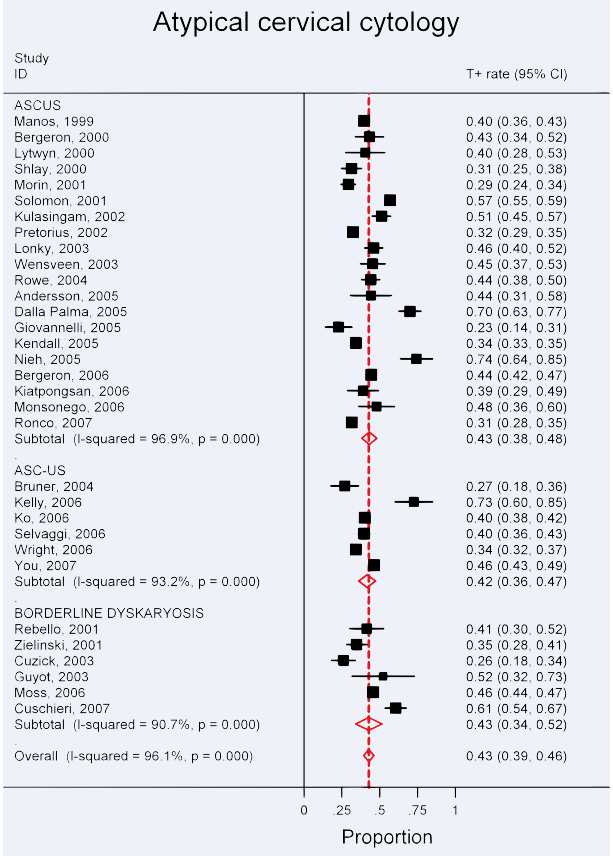

Fig. 1 Meta-analysis of the proportion of women with ASCUS or a borderline Pap smear that have a positive Hybrid Capture II test.

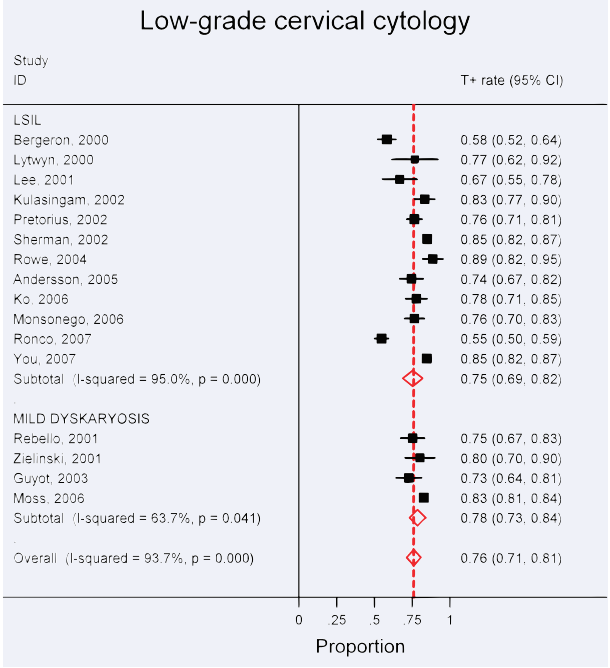

Fig. 2 Meta-analysis of the proportion of women with LSIL or a mildly dyskaryotic Pap smear that have a positive Hybrid Capture II test.

that could be included in the meta-analysis [13, 16, 36–65]. In 20 studies, high-risk HPV positivity could be derived for ASCUS defined according to TBS-1991, in six studies for AS-CUS, defined according to TBS-2001 and in six studies for borderline dyskaryosis, based on the BSCC terminology. The test-positivity rate varied between 22.8% [53] and 74.2% [55] (see Fig. 1). In spite of the wide and statistically significant inter-study heterogeneity, the pooled HPV positivity rates did not differ significantly by used cytological classification system (43.1% in ASCUS; 41.6% in ASC-US and 42.8% in borderline dyskaryosis; P for inter-group heterogeneity = 0.75). The overall pooled test positivity was 42.8% (95% CI: 39.5–46.1%).

Sixteen LSIL/mild dyskaryosis triage studies could be included in the meta-analysis enrolling 5,389 women [13, 17, 37, 38, 41–44, 46, 50, 51, 59, 60, 64–66]. In 12 and 4 studies, respectively, TBS and the BSCC were used. The lowest HC2-positivity rate was reported by Ronco (54.6%) [64] and the highest by Rowe (88.6%) [50] (see Fig. 2). The range of variation (highest–lowest proportion) in the low-grade group was smaller (34.0%) than in the ASCUS/borderline group (51.4%) but the inter-study heterogeneity was statistically significant in both meta-analyses (P for Cochrane's Q-test <0.001). The pooled hrHPV positivity rate did not significantly vary by cytological classification system used: 75.3% in LSIL and 78.5% in mild dyskaryosis; $P = 0.32$). The overall pooled HC2 positivity rate was 75.9% (95% CI: 71.2–80.6%).

The difference in the HC2-positivity rate between ASCUS/borderline dyskaryosis and LSIL/mild dyskaryosis always was positive and significantly different from zero in all studies except in one, where it approached significance (Fig. 3) [46]. The overall difference in positivity rate, pooled from 15 studies, was 32.2% (95% CI: 26.8–37.7%).

In 11 studies, age-specific results were provided (Table 1) [13, 17, 39, 49, 50, 54, 56, 61, 62, 64, 67]. However, no subgroup meta-analysis could be performed since the definition of the age strata was not uniform. A consistent and statistically significant negative trend with increasing age was observed in both triage groups (P value for trend test always <0.01). In general, hrHPV rates were higher than 80% among women younger than 30 years, with LSIL/mild dyskaryosis, at the exception of Italy, where 72% of women younger than 35 years were HPV positive. In a triage pilot project, conducted in the UK, a rate of 51% (95% CI: 41.9–60.6%) was observed only in women of 50 years and older with mild dyskaryosis [13].

Table 1 Change by age group in the HPV test positivity rate (HC2 Assay, high-risk probe, signal>1 pg/mL) in the ASCUS/borderline dyskaryosis group and LSIL/mild dyskaryosis group

ASCUS/ borderline dyskaryosis					*LSIL/mild dyskaryosis*		
Study	**Age group (years)**	***N***	**T^+**	**T^+ rate**	**N**	**T^+**	**T^+ rate**
Shlay, 2000	<30	76	37	48.7%	-	-	-
	≥30	119	24	20.2%	-	-	-
P (chi-square trend) < 0.001							
Sherman, 2002	18–22	701	498	71.0%	383	332	86.7%
	23–28	653	426	65.2%	299	263	88.0%
	≥29	844	263	31.2%	166	124	74.7%
P (chi-square trend) < 0.001					*P* (chi-squre trend) = 0.007		
Bruner, 2004	40–49	54	13	24.1%	-	-	-
	50–59	27	6	22.2%	-	-	-
	60–69	11	6	54.5%	-	-	-
	≥70	1	0	0.0%	-	-	-
P (chi-square trend) = 0.405							
Rowe, 2004	≤25	446	244	54.7%	-	-	-
	26–40	372	134	36.0%	-	-	-
	41–50	224	30	13.4%	-	-	-
	>50	243	30	12.3%	-	-	-
P (chi-square trend) < 0.001							
Boardman, 2005	<20	94	72	76.6%	-	-	-
	20–25	231	167	72.3%	-	-	-
	>25	202	118	58.4%	-	-	-
P (chi-square trend) = 0.115							
Kendall, 2005	≤20	1064	622	58.5%	-	-	-
	21–25	1800	909	50.5%	-	-	-
	26–30	1049	385	36.7%	-	-	-
	31–35	828	207	25.0%	-	-	-
	36–40	847	138	16.3%	-	-	-
	41–45	646	84	13.0%	-	-	-
	46–50	462	62	13.4%	-	-	-
	51–55	252	40	15.9%	-	-	-
	56–60	169	20	11.8%	-	-	-
	>60	217	34	15.7%	-	-	-
P (chi-square trend) < 0.001							
Bergeron, 2006	≤20	208	104	50.0%	-	-	-
	21–25	473	262	55.4%	-	-	-
	26–30	465	238	51.2%	-	-	-
	31–35	506	229	45.3%	-	-	-
	36–40	403	150	37.2%	-	-	-
	41–45	415	128	30.8%	-	-	-
	46–50	298	62	20.8%	-	-	-
	51–55	174	50	28.7%	-	-	-
	56–60	60	13	21.7%	-	-	-
	>60	45	18	40.0%	-	-	-

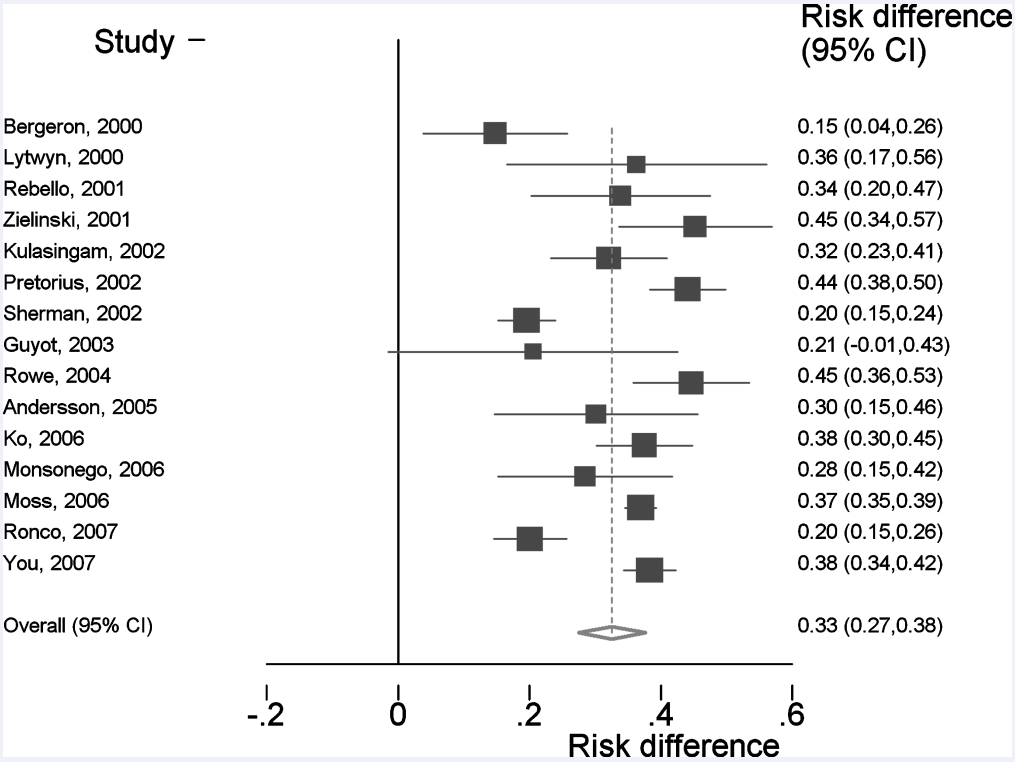

Fig. 3 Difference in Hybrid Capture 2 test-positivity rate between LSIL/mild dyskaryosis and ASCUS/borderline dyskaryosis cases.

Two studies defined HPV positivity at the cut-off of 0.2 pg of HPV DNA per millilitre and were therefore not included in the general meta-analysis shown in the Figs 1 and 2 [68, 69]. Nevertheless, the data of these studies and those from other included studies that provided values for higher thresholds [17, 46, 64] were included in the meta-regression. The HPV test positivity rate was 1.60 times higher (95% CI: 1.50–1.68%) in the LSIL/mild dyskaryosis group compared to the ASCUS/borderline group (see Table 2). The positivity rate decreased when the cutoff increased (–3.9%; 95% CI: –2.0 to –5.8% per additional RLU unit) ($P < 0.001$). The HPV positivity did not vary significantly by continent in ASCUS/borderline smears ($P > 0.20$); however, in LSIL/mild dyskaryisis, rates were significantly lower in European studies compared to American studies (RR = 0.83; 95% CI: 0.71–0.94%). There was no significant trend by year of publication ($P = 0.33$). The method of collection of cellular material used for HPV testing (Standard Transport Medium, ThinPrep [Cytyc Corporation, Boxborough, MA, USA], BD-SurePath [TriPath Imaging Inc., Burlington, NC, USA)], other and undefined methods) was not significant either. The test positivity was not significantly different in studies with complete and incomplete verification, added to the meta-analysis because of availability of age details ($P = 0.36$).

Discussion

Substantially higher HPV rates were observed in women with LSIL than among women with ASCUS. This high rate compromises the clinical utility of HPV triaging of LSIL. In the ALTS trial, further enrolment of women with LSIL was interrupted early, when preliminary analyses revealed a HC2-positivity rate of 83% [70, 71]. Testing for high-risk HPV at 12 months after the first observation of LSIL and a negative colposcopy yielded a sensitivity to predict subsequent CIN2 or CIN3 of 92% while referring 55% for repeat colposcopy [72].

The American Society for Colposcopy and Cervical Pathology (ASCCP) recommends that women with low-grade intraepithelial lesions in cytology should be referred to colposcopy [10]. It recommends that large loop excision of the transformation zone is acceptable only when CIN2 or CIN3 is found in histopathology. When colposcopy and biopsies are normal or reveal only CIN1, HPV testing after 12 months is recommended [3, 73]. The ASCCP did not recommend reflex HPV testing in case of LSIL because of the excessive hrHPV test-positivity rates observed in the ALTS study. The pooled results of our meta-analysis are consistent with the ALTS findings.

Table 1 Continued

P (chi-square trend) < 0.001							
Moss, 2006	20–34	1924	1239	64.4%	1335	1188	89.0%
	35–49	1217	353	29.0%	373	259	69.4%
	50–64	543	88	16.2%	117	60	51.3%
P (chi-square trend) < 0.001					*P* (chi-square trend) < 0.001		
Selvaggi, 2006	<20	57	32	56.1%	-	-	-
	21–25	189	94	49.7%	-	-	-
	26–30	159	61	38.4%	-	-	-
	31–35	91	34	37.4%	-	-	-
	36–40	30	9	30.0%	-	-	-
	41–45	32	8	25.0%	-	-	-
	46–50	41	11	26.8%	-	-	-
	51–55	30	8	26.7%	-	-	-
	56–60	22	5	22.7%	-	-	-
	>60	21	4	19.0%	-	-	-
P (chi-square trend) < 0.001							
Wright, 2006	≤25	446	244	54.7%	-	-	-
	26–40	372	134	36.0%	-	-	-
	41–50	224	30	13.4%	-	-	-
	>50	243	30	12.3%	-	-	-
P (chi-square trend) < 0.001							
Ronco, 2007	<35	241	110	45.6%	219	157	71.7%
	≥35	516	128	24.8%	266	108	40.6%
P (chi-square trend) = 0.001					*P* (chi-square trend) = 0.002		

A British pilot study also showed that the great majority of women with mild dyskaryosis were infected with high-risk papillomavirusses, precluding efficient triage by general hrHPV DNA testing. Eighty-four percent of women with mild dyskaryosis had a positive HC2 result. In women younger than 35 years, the test positivity rate even reached 89% [13]. To reduce the excessive number of colposcopies, the triage policy was revised in two of the three participating laboratories and women younger than 35 years were referred only when, 6 months later, they remained HPV positive or showed mild dyskaryosis or worse. The option to postpone HPV triage in women with mild dyskaryosis aged 35 or older was not explored nor simulated in the cost-effect analysis, in spite of the fact that seven to eight out of ten were hrHPV positive [74]. However, in The Netherlands, Bais and Berkhof showed that delayed HPV and repeat cytology testing in patients with borderline or mild dyskaryosis after 6 and 18 months is both safe and more cost-effective than immediate HPV triage [75, 76]. Postponing triage, allows viral clearance, which over a period of 6 to 12 months can vary from 18% to 45% [75] and therefore reduces the need for colposcopy.

From a recent Italian trial, it was suggested that hrHPV triage could be useful in women with LSIL above 35 years of age [64]. However, in this study, rates in hrHPV rates were out-lying (lowest of all studies and 22% lower than the pooled average).

In the case of borderline lesions, where HPV test positivity ranges, on average, between 40% and 50%, the number of colposcopies can be reduced considerably by virological triage. Nevertheless, in women younger than 30–35 years, the specificity and the positive predictive value are low as well [9, 62, 67, 77].

Management of women with minor cytological cervical lesions could be made more specific by increasing the cut-off of a positive HC2 test, but to date insufficient data are available to allow definite conclusions on whether raising of cut-off can be done without compromising sensitivity.

Recent data indicate that typing for the most oncogenic HPV types, in particular for HPV16, has the potential to select women with minor cytological lesions that have a high risk for having or developing CIN3 [78–80]. The ROC curve, in Fig. 4,

Table 2 Relative risk of hrHPV positivity computed from a metaregression

All minor lesions	RR	lcib	ucib
Cytological category			
Atypical cervical cytology (= reference)	1		
Low-grade cervical cytology	1.60	1.50	1.68
Year of publication (ref = 1998)	0.99	0.96	1.01
RLU	0.96	0.94	0.98
Atypical cervical cytology			
Cytological subcategory			
ASCUS (= reference)	1		
ASC-US	0.95	0.67	1.25
Borderline dyskaryosis	1.03	0.74	1.35
Continent			
America (= reference)	1		
Asia	1.26	0.87	1.64
Europe	0.97	0.72	1.24
Low-grade cervical cytology			
Cytological subcategory			
LSIL (= reference)	1.00		
Mild dyskaryosis	1.10	1.01	1.15
Continent			
America (= reference)	1		
Asia	0.96	0.82	1.06
Europe	0.83	0.71	0.94

ASCUS: atypical squamous cell of undetermined significance (defined according to 1992 version of The Bethesda System); ASC-US: atypical squamous cell of undetermined significance (defined according to 2002 version of The Bethesda System); LSIL: low-grade squamous intraepithelial lesion; RLU: relative light units (relative unit to express viral load); RR: relative risk; lcib: lower 95% confidence interval bound; ucib: upper 95% confidence interval bound.

illustrates the change in the trade-off in sensitivity and specificity (using $CIN3^+$ as outcome) by targeting increasingly more HPV types using ASCUS triage data from the ALTS study [78]. At each step, the type was chosen that yielded the largest gain in sensitivity and the lowest loss in specificity when the gain in sensitivity was equal for more types. HPV16 was the first type to be included, showing a sensitivity of 56% for a specificity of 89%. Adding HPV31 improved the sensitivity by 8% and decreased the specificity by 5%. The number of triage positive women increased considerably by testing for more than 10 types, with only a very small additional detection of CIN3 cases. Figure 4 also contains the point corresponding with the accuracy of HC2 [8]. HC2 targets 13 high-risk types, but also cross-reacts with certain other high- and low-risk types [81]. Therefore, HC2 could reach sensitivity for $CIN3^+$ of 92% for a specificity of only 51% [8]. In the Guanacaste cohort, Castle found a 39% absolute risk for prevalent or incident CIN3 in LSIL women infected with HPV16, whereas women infected with one of the other 12 types included in the high-risk probe of the HC2 assay expressed a risk for $CIN3^+$ of only 10% [82]. These data suggest that focused management of ASCUS cases carrying HPV16 or some other types (located in the left part of the ROC curve in Fig. 4) and more conservative follow-up of ASCUS cases infected with other types (near the right part of the curve) could increase triaging efficiency.

Triage with a poorly specific test unnecessarily labels women as being at risk for cervical cancer, may induce anxiety and overtreatment and possible subsequent adverse obstetrical effects [83, 84]. Certain molecular markers of early carcinogenic transformation could make triage more specific. One small LSIL triage study, using tests for messenger-RNA for viral oncoproteins E6 or E7 with a follow-up of 2 years, showed a low-test positivity rate (27%) and a high specificity (93%) for subsequent high grade CIN [85]. Triage with $GP5^+/CP6^+$ PCR showed a positivity rate of 74%, a specificity of 29%, whereas the sensitivity (80%) was equal to that of mRNA testing. Over-expression of certain cell cycle regulator proteins, integration of HPV DNA sequences in the human genome or determination of certain genetic or immunologic profiles are potential candidates for adjunct triage testing [86–91]. A recent meta-analysis established a clear correlation between p16 over-expression and the severity of cytological lesions, however the variation of p16 positivity was extremely large (ranging between 10% and 100% in ASCUS and between 24% and 86% in LSIL), underlining lack of standardization in immuno-staining, interpretation and reporting [92]. Nevertheless, in experienced hands and using clearly defined criteria, p16 immuno-staining has shown excellent results with sensitivities for $CIN2^+$ similar to HC2, remarkably lower positivity rates (27% in ASCUS, 24% in LSIL) and consequently substantially higher specificities (84% and 81%, in ASCUS and LSIL, respectively) [93]. These promising results invite for more powerful well-designed studies to evaluate the role of p16 and other biomarkers that identify progressive cervical lesions and/or transforming HPV infections. Currently, we must acknowledge the lack of good triage studies comparing p16 with currently used alternative strategies to triage minor cytological lesions. We note only one recent Italian study, where p16-immunostaining was used in the background of HPV screening to triage HPV-positive women. P16-enhanced cytology showed a higher sensitivity and similar positive predictive value for high-grade CIN compared to non-stained conventional cytology [94]. In order to explore the potential to use p16 over-expression as a progression marker in triage, we propose to

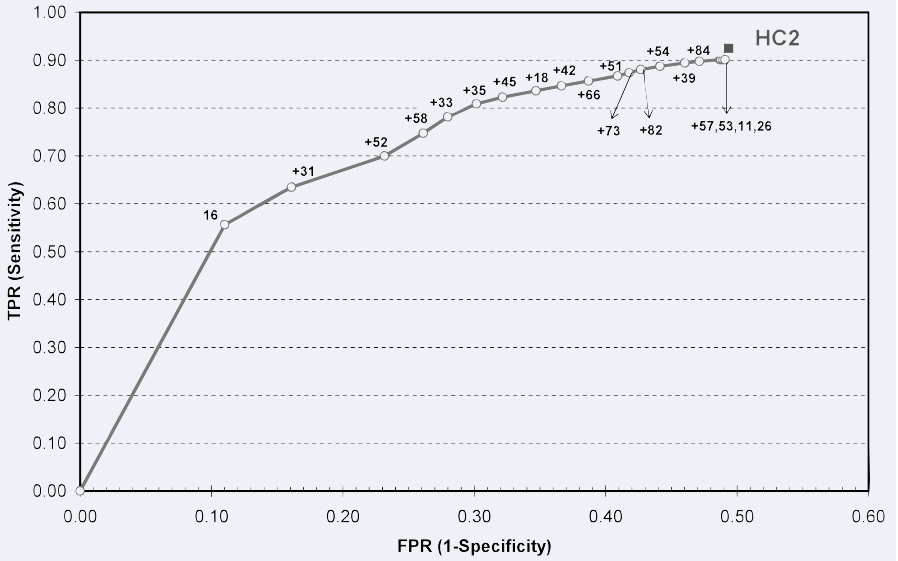

Fig. 4 ROC curve representing the true positivity rate (TPR = sensitivity) for CIN3 or worse disease as a function of the false-positivity rate (FPR = 1-specificity) for increasingly sensitive combinations of HPV genotypes. The plot is constructed using data received from the ASCUS-LSIL triage study, and concerns the accuracy of an HPV multityping assay applied on women with ASCUS [82]. The square in the upper right corner corresponds with the sensitivity and specificity for $CIN3^{+}$ of the Hybrid Capture II assay [8].

set up an international workshop to standardize issues of sample processing and to define clear criteria for categorizing levels of positivity.

Our meta-analysis also provides substance for the use of hrHPV testing with a standardized assay as quality control method in cytopathology which could become an alternative for established re-screening practices [95–97]. High-risk HPV postivity rates could be used to identify laboratories or cytotechnologists that overcall or undercall equivocal or low-grade abnormalities. If we should use the range –/+ two standard deviations around the pooled test-positivity rates derived from our meta-analysis, we could consider 25–61% as benchmark for equivocal squamous cytology and 57–95% for low-grade lesions. In that case, one study could be hypothesized as overcalling ASC-US [53], two studies as under-calling ASCUS [52, 55] and one study as over-calling LSIL [64]. Certainly, more research and debate is necessary before this idea can be translated into evidence-based guidelines.

Conclusion

Around three quarters of women with LSIL are hrHPV positive compared to less than half with equivocal cytological abnormalities. The pooled results of our meta-analysis are in agreement with ALTS findings and indicate that reflex hrHPV testing is insufficiently discriminative in case of LSIL. In older women with LSIL, hrHPV testing could be useful, but currently no obvious age threshold can be defined by lack of reported age-specific data.

More meta-analytical work is needed, based on 5-year age groups or using individual patient data, also including other HPV testing assays, to provide guidance on LSIL triage. For this purpose, we are currently obtaining age-stratified data from published triage studies. This effort generating clinically relevant evidence will require collaboration between systematic reviewers and the principal investigators of published or ongoing studies [98].

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