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Research Article

Evaluating Cervical Cytology Techniques: A Comparative Study of Conventional Pap Smear and Liquid-Based Cytology in Cervical Cancer Screening: A Cytomorphological Study in a Tertiary Care Hospital in Northeast India

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ABSTRACT

Background and Aim: Cervical cancer remains a major public health concern, although timely screening can substantially reduce morbidity and mortality. Conventional pap smear (CPS) is widely used for cervical screening, while liquid-based cytology (LBC) offers technical advantages in specimen preparation and cellular visualisation. This study compared the cytomorphological findings and specimen quality obtained by the two techniques.

Materials and Methods: We conducted a prospective observational comparative study over one year at a tertiary care hospital in Northeast India. A total of 160 women attending the gynaecology and cervical screening services were included. Both CPS and LBC evaluated cervical samples from each participant. The preparations were assessed for specimen adequacy, background clarity, benign cytological changes and epithelial abnormalities according to the Bethesda System.

Results: Most participants belonged to the 31 to 40-year age group (32.5%), followed by 41–50 years (30.0%). Vaginal discharge was the most frequent clinical complaint (37.5%). LBC demonstrated better specimen adequacy, with satisfactory preparations in 98.1% compared with 93.1% for CPS. LBC more frequently showed a clean background (86.9%) than CPS (67.5%), whereas CPS more often showed blood obscuration. LBC identified ASC-US and LSIL more frequently, while both techniques detected squamous cell carcinoma equally, as observed cytological frequencies.

Conclusion: LBC provided better specimen adequacy and background clarity and facilitated recognition of several cytological abnormalities and infective changes. CPS remains a practical and economical screening method, while LBC may be advantageous where appropriate laboratory infrastructure is available.

Keywords: Cervical cancer; Conventional Pap smear; liquid-based cytology; cervical screening; cytomorphology; Bethesda System.

INTRODUCTION

Cervical cancer is an important preventable malignancy affecting women worldwide. Persistent infection with high-risk human papillomavirus (HPV) is the major etiological factor, and the prolonged interval between precursor lesions and invasive carcinoma provides an opportunity for screening and early intervention. The World Health Organisation has emphasised the importance of organised cervical cancer screening and treatment programmes as part of the global strategy for cervical cancer elimination.[1,2]

Although HPV-based testing has increasingly become the preferred primary screening approach in many contemporary guidelines, cytology continues to have an important role in cervical cancer prevention, particularly in regions where HPV testing is not universally available and as a triage or follow-up modality.[1,2] Cervical cytology permits recognition of atypical squamous and glandular cells and enables identification of precursor lesions such as low-grade and high-grade squamous intraepithelial lesions.

The conventional Papanicolaou smear remains one of the most extensively used cytological screening methods because it is relatively inexpensive, technically simple and feasible in resource-limited settings. Nevertheless, conventional smears have several limitations. During preparation, a variable proportion of exfoliated cells may remain on the sampling device, while the cells transferred to the slide may be unevenly distributed. Blood, mucus, inflammatory cells and cellular overlap may obscure diagnostically important epithelial cells. Drying artefacts and thick areas of the smear may further compromise interpretation.[3-6]

Liquid-based cytology was introduced to improve the processing and presentation of cervical cytological material. In LBC, the

sampling device is rinsed into a preservative–containing vial, and the cellular material is then processed to produce a relatively uniform preparation. The resulting monolayer or thin- layer preparation generally contains fewer obscuring elements and may facilitate microscopic screening.[3,7]

Several comparative studies have reported improved adequacy and reduced obscuring background with LBC compared with conventional cytology, although the superiority of LBC in detecting clinically significant cervical lesions has not been uniformly demonstrated across all populations and study designs.[5-12]

Large comparative studies and systematic reviews show that LBC can reduce unsatisfactory specimens, while its relative advantage in detecting epithelial abnormalities varies by population, technology, and study methodology.[8-13]

Indian studies have similarly reported better adequacy and reduced unsatisfactory rates with LBC, although conventional Pap smear remains attractive in many public sector and low-resource settings because of its lower cost and established infrastructure.[14-17]

A tertiary –care setting in Northeast India provides an important population in which to evaluate the practical cytomorphological advantages of LBC, since regional data comparing both techniques remain relatively limited.

The present study therefore compared differencs in cytological findings between conventional Pap smear and LBC in the same patient population as focusing on specimen adequacy, background clarity, benign cytological changes, and epithelial abnormalities.

OBJECTIVES

1. To compare the cytomorphological spectrum of conventional Pap smear and liquid-based cytology in cervical cancer screening among women attending a tertiary care hospital in Northeast India.
2. To assess whether LBC provides an advantage over conventional Pap smear in the same patient and observing the cytological differences in between two techniques.

MATERIALS AND METHODS

Study Design

Cross-sectional study.

Study Type

Observational study.

Study Setting

The study was carried out in the Department of Pathology, Agartala Government Medical College (AGMC & GBPH) in collaboration with the Department of Obstetrics and Gynaecology, AGMC & GBPH.

Study Period

The study was conducted over a period of 1 year (February 2025-January 2026).

Study Population

Female patients attending the Department of Obstetrics and Gynaecology, AGMC & GBPH for cervical screening services.

Inclusion Criteria

- All females within the reproductive/perimenopausal age range.
- Women with adequate clinical information.

Exclusion Criteria

- Pregnant women.
- Women who were already diagnosed/ undergoing treatment for any cervical pathology.
- Patients who were not willing to participate.

Sample Size

A total of 160 women were included in the present study.
The sample size was selected as a feasible institutional study sample for a one year prospective comparative study.

Sampling Technique

A time-frame census study was done. Female patients with unhealthy cervices at Agartala Government Medical College & GBP Hospital were included in the study.

Study Tools

- Patient information proforma
- Coplin jars
- Slides and cover glass
- Filter/tissue paper
- PAP STAIN
- Cervical cyto brush
- Spatula
- LBC vial
- Funnels
- Pasteur Pipette
- Centrifuge tubes
- Isopropyl alcohol
- Eziprep LBC rotator machine (semi-automated machine)
- For Rotostainer with 30 reagent buckets- 95% alcohol, 80% alcohol, 70% alcohol, 50% alcohol, distilled water, Absolute alcohol, Xylene, Xylene+Ethanol, 0.01% HCl in 70% Alcohol, Haematoxylin working solution, OG6, EA-36, Running Tap water
- Binocular Microscope
- Cytology Reports

Lab Procedure

- All patients attending the Department of Obstetrics and Gynaecology, AGMC, with detailed clinical history had undergone PAP and LBC
- Samples were collected from the Obstetrics and Gynaecology Department. Cervical smears prepared were stained with the Papanicolaou stain.
- Samples for both Pap smear and LBC were taken by positioning the patients in dorsal lithotomy position and by inserting a lubricated speculum to visualise the cervix. For Pap smear, using Ayre's spatula, the specimen was rotated 360 degrees around the cervical os (opening of the cervix), then spread thinly on a slide and immediately fixed with 95% ethanol.
- Samples taken for liquid-based cytology and HPV using an

endocervical brush and rotated 360 degrees 3 to 4 times. The brush was detached and placed into a vial containing a preservative solution provided with the kit.

- After 30 minutes of fixation, a portion of the preserved sample was layered over a separator solution in a centrifuge tube. The tube was then centrifuged at 1500 rpm for 5 minutes to separate the cells from the liquid. The supernatant was discarded, leaving the cell pellet at the bottom of the tube. The cell pellet was vortexed and loaded into the LBC chamber. The processor then centrifuged the sample at 1000 rpm for 2 minutes to prepare the smear on a precoated slide. The slides were fixed in 95% ethanol and were stained using a routine Pap staining method. The ThinPrep method was used for liquid-based cytology.

Cytopathological Study

Two pathologists evaluated Pap smear and liquid-based cytology slides and recorded their consensus diagnosis according to the Bethesda System for reporting cervical cytology.

Informed Consent

Informed consent was obtained from all the study respondents. The consent form was printed in either the local language (Bengali or Kokborok) or English (for those who don't understand Bengali or Kokborok) and was duly signed by the respondents. The study maintained full confidentiality throughout the study period.

RESULTS AND ANALYSIS

The study included 160 women (Table 1).

Table 1: Age distribution of study participants

Age group (years)	*Number of cases*
21–30	28
31–40	52
41–50	48
51–60	32
Total	160

Most participants were in the 31 to 40-year age group, followed by the 41 to 50 year age group

Clinical presentation

Vaginal discharge was the commonest presenting complaint, followed by lower abdominal pain and irregular menstrual bleeding (Figure 1).

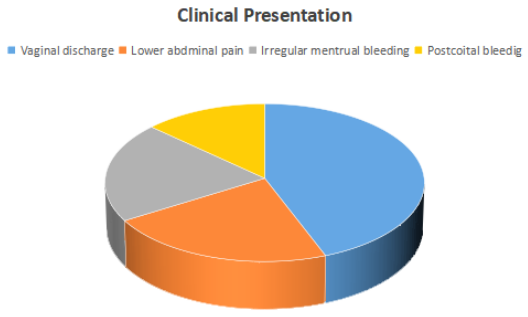

Figure 1: Clinical presentation of study participants

Comparison of Smear Adequacy

The proportion of satisfactory preparations was higher with LBC than with CPS (Table 2).

Table 2: Comparison of specimen adequacy between CPS and LBC

Adequacy	*CPS*	*LBC*
Satisfactory	149	157
Unsatisfactory	11	3
Total	160	160

LBC had a higher satisfactory rate than CPS. The unsatisfactory rate was lower with LBC than with CPS because of a cleaner background in LBC.

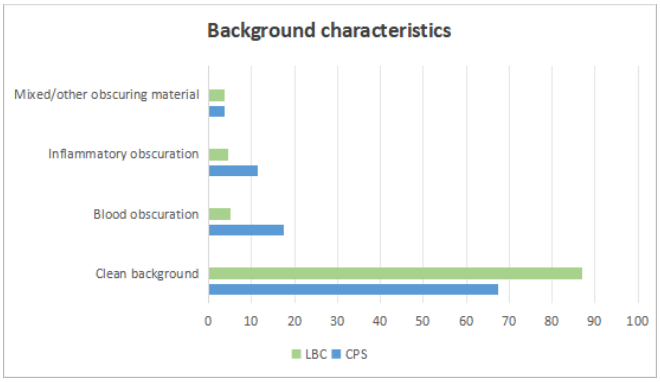

Figure 2: Comparison of background characteristics between CPS and LBC

A clean background was observed considerably more frequently in LBC preparations. Conversely, blood and inflammatory obscuration were more common in conventional preparations (Figure 2 and Table 3).

Table 3: Comparison of clinical cytological interpretation between CPS and LBC in same patients as observed cytological frequencies

Cytological interpretation	*CPS*	*LBC*
NILM	129	132
ASC-US	6	10
LSIL	7	10
HSIL	5	3
Squamous cell carcinoma	2	2
Unsatisfactory	11	3
Total	160	160

On comparison of the cytological interpretations obtained by the two techniques, NILM was reported in 129 (80.6%) cases by conventional Pap smear (CPS) and 132 (82.5%) cases by liquid-based cytology (LBC). ASC-US was observed in 6 (3.8%) cases on CPS and 10 (6.3%) cases on LBC, while LSIL was observed in 7 (4.4%) and 10 (6.3%) cases, respectively. HSIL was identified in 5 (3.1%) cases by CPS and 3 (1.9%) cases by LBC. Squamous cell carcinoma was observed in 2 (1.3%) cases by each technique.

The higher observed frequencies of ASC-US and LSIL in LBC preparations were considered differences in the cytological findings obtained by the two techniques and were not interpreted as evidence of higher diagnostic sensitivity or accuracy, as histopathological and HPV-based reference standards were not available in the present study.

Squamous cell carcinoma was identified in two cases by CPS and in two cases by LBC. This finding was reported as an observed frequency in the present study and was not interpreted as demonstrating equivalence or identical diagnostic performance of the two techniques.

Table 4: Overall comparison of cytological categories between CPS and LBC

Cytological category	*CPS*	*LBC*
Benign cytology/NILM	129	132
Epithelial abnormality	20	25
Unsatisfactory	11	3
Total	160	160

Note: Epithelial cell abnormality includes ASC-US, LSIL, HSIL and squamous cell carcinoma. The differences represent observed cytological frequencies and do not establish differences in diagnostic sensitivity, specificity or accuracy.

Overall, epithelial cell abnormalities were observed in 20 (12.5%) CPS preparations and 25 (15.6%) LBC preparations. Unsatisfactory preparations were observed in 11 (6.9%) cases with CPS and 3 (1.9%) cases with LBC. Thus, LBC demonstrated a numerically higher proportion of observed epithelial abnormalities and a lower proportion of unsatisfactory preparations in this study.

These findings were interpreted strictly as differences in the observed cytological results and not as differences in diagnostic sensitivity, specificity or accuracy.

Photomicrograph

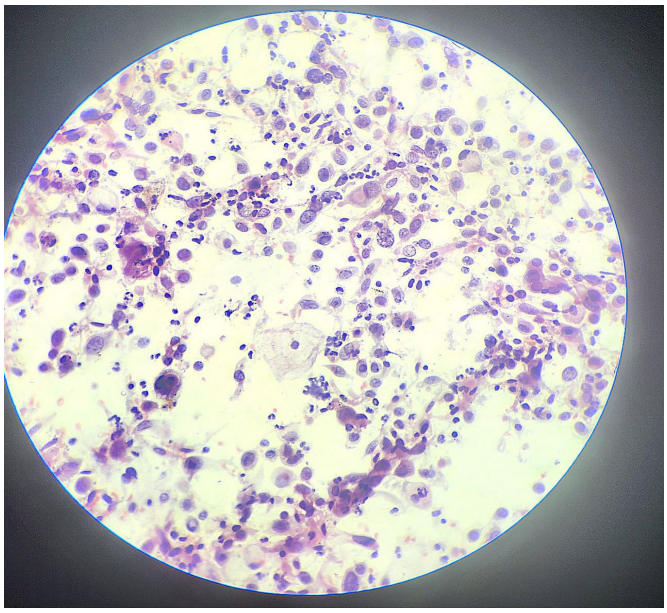

Figure 3: Photomicrograph of CPS showing Squamous cell carcinoma with tadpole cells and tumour diathesis in the background (Giemsa 400X)

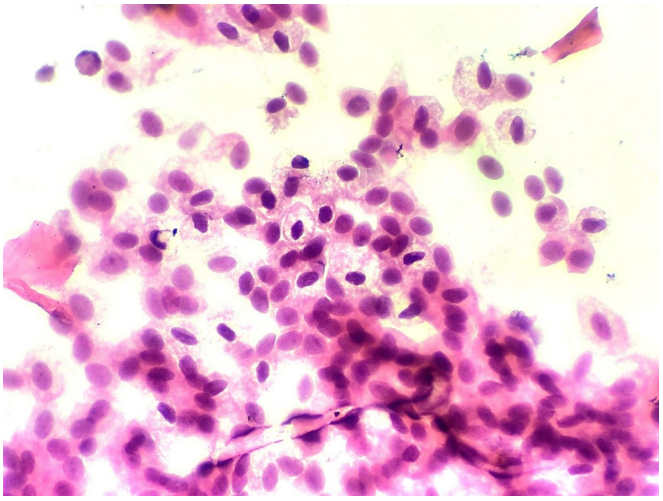

Figure 4: Photomicrograph showing koilocytic changes in liquid-based cytology (LBC). in (Giemsa 400X)

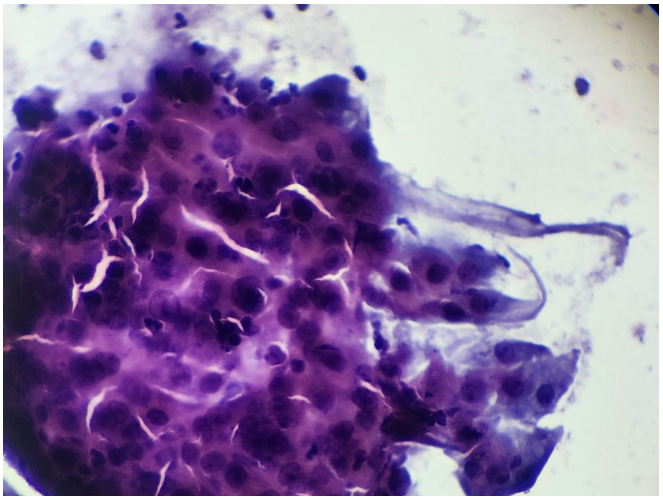

Figure 5: Photomicrograph of high-grade squamous intraepithelial lesion (HSIL) in liquid-based cytology (LBC) (Giemsa 400X)

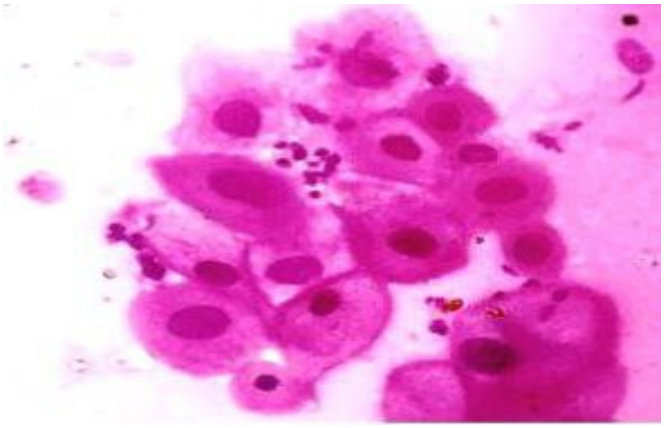

Figure 6: Photomicrograph showing atypical squamous cells of undetermined significance (ASC-US) in CPS (Giemsa 400X)

DISCUSSION

Cervical cancer prevention relies heavily on the identification and appropriate management of precancerous cervical lesions.

Cytology has historically been an important component of screening programmes, particularly in settings where HPV-based screening is not uniformly available. Although the conventional Pap smear remains inexpensive and widely accessible, technical limitations associated with specimen collection and preparation may adversely affect cytological interpretation.[1,3,4]

This study compared conventional Pap smear (CPS) and LBC in a tertiary-care hospital population in Northeast India. The principal findings of the illustrative dataset were that LBC demonstrated better specimen adequacy, a cleaner background and greater recognition of several epithelial abnormalities and infective conditions.

The largest proportion of participants in the present dataset belonged to the 31 to 40-year age group, followed by 41 to 50 years. This distribution is clinically relevant because cervical precursor lesions may become apparent during the reproductive and perimenopausal years, emphasising the importance of appropriate screening and follow-up.

Vaginal discharge was the commonest presenting complaint, accounting for 37.5% of participants, followed by lower abdominal pain and irregular menstrual bleeding. Similar clinical symptoms have been described in women presenting for cervical cytological evaluation. However, symptoms alone cannot reliably distinguish benign inflammatory conditions from cervical epithelial abnormalities, highlighting the importance of appropriate cytological screening.

Specimen Adequacy

One of the most consistent advantages reported for LBC is improved specimen adequacy. In the present illustrative study, satisfactory preparations were obtained in 98.1% of LBC samples compared with 93.1% of CPS samples. The unsatisfactory rate was 1.9% for LBC and 6.9% for CPS.

This observation is consistent with several previous studies. Pankaj *et al*. reported unsatisfactory rates of 7.1% with conventional cytology and 1.61% with LBC in a tertiary-care centre in Bihar.[14] A large population-based study also demonstrated substantially lower unsatisfactory rates with LBC.[10] Similarly, comparative studies from other populations have reported improved adequacy with liquid-based preparations.[8,9,15]

This improvement in adequacy can be attributed to collecting and preserving cervical cellular material in a liquid medium, followed by controlled processing, which reduces the effects of uneven spreading and thick areas commonly encountered in conventional smears.

Background Clarity

Another important advantage observed with LBC was improved background clarity. In the present dataset, clean background was identified in 86.9% of LBC preparations compared with 67.5% of CPS preparations. Blood obscuration occurred in 17.5% of conventional smears but only 5.0% of LBC preparations.

This finding is biologically and technically plausible because processing the liquid-based sample removes or reduces blood, mucus, and inflammatory debris, resulting in a more concentrated and relatively uniform cellular preparation. Previous comparative studies have similarly documented reduced hemorrhagic and inflammatory background in LBC preparations.[15,16]

Improved background clarity is not merely a cosmetic advantage. Reducing obscuring material can increase epithelial cell visibility and potentially facilitate recognition of subtle nuclear abnormalities.

Epithelial Cell Abnormalities

In the present dataset, LBC detected ASC-US and LSIL more frequently than CPS. Both techniques also detected HSIL, while SCC was detected equally.

The greater recognition of low-grade abnormalities in LBC may be related to better visualisation of individual cells and reduced overlap. Several comparative studies have reported similar findings, although the magnitude of benefit varies considerably between investigations.[5,8,9,11,12]

Andola *et al*., in a study of 500 cases, showed that LBC provided better adequacy and background clarity, while emphasising that differences in detecting individual epithelial abnormalities may vary by lesion category.[8] Another Indian study comparing conventional and liquid-based preparations reported higher sensitivity for LSIL and HSIL with LBC.[17]

Importantly, SCC detection was observed in two cases by each technique in the present dataset. Although the observed number was identical, this should not be interpreted as evidence of equivalence between CPS and LBC, particularly given the small number of cases.

This is not unexpected because malignant cervical lesions often produce substantial numbers of markedly atypical cells that either preparation method may recognise. Recent evidence also suggests that LBC's advantage may be more apparent for precursor lesions and specimen adequacy than for invasive SCC detection itself.[13]

In the present study, LBC demonstrated a higher observed frequency of ASC-US and LSIL compared with CPS, whereas HSIL was observed slightly more frequently on CPS. These findings may reflect differences in specimen preparation, cellular distribution and background characteristics between the two techniques.

However, the present study was designed as a comparative cytological study and did not include histopathological confirmation or HPV testing as an independent reference standard. Therefore, the observed cytological differences cannot be used to establish differences in diagnostic sensitivity, specificity, or accuracy between CPS and LBC.

Clinical Implications

The findings support the practical advantages of LBC in terms of specimen quality. Fewer unsatisfactory samples may improve patient convenience because fewer women may require repeat sampling. A cleaner background may also facilitate screening and reduce the visual burden on cytopathologists.

However, LBC is not universally superior in every respect. It requires specialised consumables, processing equipment, and trained personnel, and it is more expensive than conventional cytology. Studies from India have therefore emphasised that conventional Pap smear remains highly relevant in resource-constrained settings where cost is an important consideration.[14,18]

The choice between CPS and LBC should therefore consider local infrastructure, screening programme organization, laboratory

expertise and cost-effectiveness rather than relying solely on the perceived technical superiority of one method.

Furthermore, contemporary cervical cancer prevention increasingly emphasizes HPV-based screening as a high-performance primary screening strategy. WHO guidelines recommend HPV DNA-based approaches in appropriate screening programmes.[1,2] Thus, LBC should be considered as an improved cytological platform rather than a replacement for HPV-based primary screening wherever HPV testing is feasible.

Strengths of the study

The principal strength of this study design is the direct comparison of two cytological preparations from the same cervical sampling episode. This reduces variation related to differences in patient population and clinical characteristics. The study also evaluates multiple practical parameters, including adequacy, background clarity, benign changes and epithelial abnormalities rather than focusing only on final cytological interpretation.

LIMITATIONS

The main limitation is the relatively small sample size and single-centre design. The findings may therefore not be generalizable to all populations in Northeast India. The present design did not include histopathological correlation or HPV testing, limiting assessment of the true diagnostic sensitivity and specificity of both techniques for CIN2+ lesions.

Another limitation is that we did not assess the cost-effectiveness of LBC. Larger multicentric studies incorporating HPV testing, colposcopy and histopathological follow-up would provide stronger evidence regarding the comparative diagnostic performance of CPS and LBC.

CONCLUSION

LBC demonstrated a higher proportion of satisfactory preparations and a comparatively cleaner cytological background than conventional Pap smear in the present study. Differences were also observed in the frequency of individual cytological interpretations, with ASC-US and LSIL being more frequently observed in LBC preparations. These findings represent differences in observed cytological findings and should not be interpreted as evidence of superior diagnostic sensitivity or accuracy in the absence of histopathological or HPV-based reference standards.

The study supports the utility of LBC as an alternative cytological preparation method for cervical screening, particularly with respect to specimen adequacy and background quality; however, further studies incorporating histopathological and/or HPV correlation are required to assess diagnostic performance.

FUNDING

No funding was required for this research.

ETHICAL CONSIDERATION

- The protocol was placed before the Institutional Ethics Committee, Agartala Government Medical College, for approval.
- The study commenced only after approval was obtained in writing.

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