Association of *Schistosoma haematobium* and Human Papillomavirus in Cervical Cancer

A Case Report

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Background

The association between Schistosoma haematobium *and cervical cancer has been reported for a long time. However, recently human papillomavirus, a cofactor in the genesis of cervical cancer, has been confirmed. A case of squamous intraepithelial lesion after* S haematobium *infection is presented, and the relation between schistosomiasis, human papillomavirus and squamous intraepithelial lesion, with long-term follow-up by Papanicolaou smear, is discussed.*

Case

A 33-year-old, normal, healthy woman with a history of Copper intrauterine device (IUD) use for 3.9 years presented for her annual contraceptive follow-up. Her Pap smear revealed inflammation with a S haematobium *egg. She was followed up with Pap smears for 4 years. Retrospective contraceptive history revealed use of a copper IUD on 5 occasions with a total duration of 13 years and 1 month. Similarly, annual follow-up of Pap smears for the past 13 years showed mild inflammation with bacterial vaginitis and monilial infection. Subsequent smears showed an* Actinomyces*-like organism and then human papillomavirus infection with atypical squamous cells of undetermined significance followed by human papillomavirus–associated low/high grade squamous intraepithelial lesion.*

Conclusion

Caution is required while screening routine Pap smears. Apart from nuclear abnormalities, one can observe unusual findings. Long-term follow-up by Pap smear following detection of S haematobium *revealed that in the absence of human papillomavirus,* S haematobium *alone is not the causative agent for the abnormal proliferation of squamous epithelium of the cervix. Genital* Schistosomia *acts as a cofactor by traumatizing the genital epithelium or immune suppression to favor human papillomavirus infection.* (Acta Cytol 2010;54:205–208)

Keywords: cervical cancer, human papillomavirus, Papanicolaou smear.

One must be aware of rare parasites while screening routine Pap smears.

Schistosoma worms are trematode parasites that inhabit the mesenteric, portal vesical and pelvic venous plexuses. Human schistomiasis is usually attributed to 6 species *Schistosoma haematobium*: *mansoni*, *japonicum*, *intercalatum*, *malayensis* and *mekongi*; the clinical features depend on the species, development stage and site of infection. Three major syndromes identified are (1) cercarial dermatitis, (2) acute schistosomiasis, or katayama fever, and (3) chronic fibroobstructive disease. *S haematobium* is endemic in 53 countries: the Middle East, African countries, in some parts of South America, Caribbean areas and an ill-defined focus in India.[1-3] It is an important health issue as the International Agency for Research on Cancer considers *S haematobium* infection a definitive cause of urinary bladder cancer, with an associated 5-fold increased risk[4] and with Indian reports providing evidence for the same assertion.[5]

For *Schistosoma*, the human being is the ultimate host, while the snail is the intermediate host. Schistosomiasis is caused when free-swimming *Schistosoma* embryos or cercariae penetrate intact skin on the epithelial surface. They mature into adult tramatodes, or flukes. *S haematobium* adults reside in the vesicle and pelvic plexuses of the venous circulation. The adult female can contain 20–100 eggs at one time. In addition to the vesicle and pelvic plexuses, ova may occur in the rectal venules. For schistosomiasis ova, the bladder is the site wherein most of the ova are released into the urine; those remaining die and incite a granulomatous tissue reaction with subsequent fibrosis. Excreted eggs develop into miracidiae, which infect freshwater snails (the intermediate host). The snails release cercariae, thereby completing the cycle.[6] The rich network of venous anastomosis between the bladder and the genital tract offers an explanation for the more common finding of *S haematobium* in the female genital tract. In addition, *S haematobium* ova may also be present in the female genital tract due to contamination since urinary tract and genital tract openings are in the vagina. Youssef et al,[7] in 1962, first reported the detection of schistomiasis by vaginal cytology and suggested that schistosomal infection of the cervix was precancerous. Since then, several investigators have reported the association of

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cervical carcinoma and schistosomiasis. Recent reports confirm an association between human papillomavirus (HPV) infection and cervical carcinoma.

There are no previous long-term follow-up studies of schistosomiasis infection by Pap smear, including with reference to initial negative normal Pap smears for the evaluation of cervical cancer etiology. We present a case with possible association of *S haematobium* and HPV infection with cervical intraepithelial neoplasia in a long-term follow-up Pap smear study.

Case Report

A 33-year-old, normal, healthy woman, gravida 3, para 2, abortion 1, with a history of a copper intrauterine device (IUD) use for 3 years and 9 months (insertion done elsewhere), reported to the institute's family welfare clinic for a contraceptive follow-up visit. Her gynecologic examination was normal, while cytology (Pap smear) revealed inflammation with ovum of *S haematobium* (Figure 1A). Pap smears are in routine use for the screening of premalignant/malignant lesions of the cervix. They have the additional benefit of detecting multiple infections, such as parasitic, protozoan, fungal, bacterial and viral, in a single smear at no additional cost.[8,9]

The patient had registered 5 years earlier for the family planning services at the family welfare clinic of this institute. During her first visit, she complained of vaginal discharge and pruritis vulvae; clinical examination revealed a copper device in situ with endocervicitis and uterine tenderness, for which she was treated with doxyclycline (100 mg twice a day for 7 days) and povidone iodine vaginal pessaries (USP 200 mg, once daily intravaginal use for 10 days). Cytology revealed only inflammation. A year later, removal and insertion of a different IUD (Copper Safe [Cu Safe 200 Prosan International BV, the Netherlands] removed and Copper T 200 [Famy Care Limited, Daman, India] inserted,) were undertaken. The patient had no signs or symptoms, and the Pap smear was negative.

One year after insertion, the patient complained of vaginal discharge and pruritis vulvae; clinical examination revealed the IUD in situ with endocervicitis and uterine tenderness, for which she was treated with doxycycline (100 mg twice a day for 14 days) and tinidazole (500 mg twice a day for 5 days); her husband was given a 1-g,

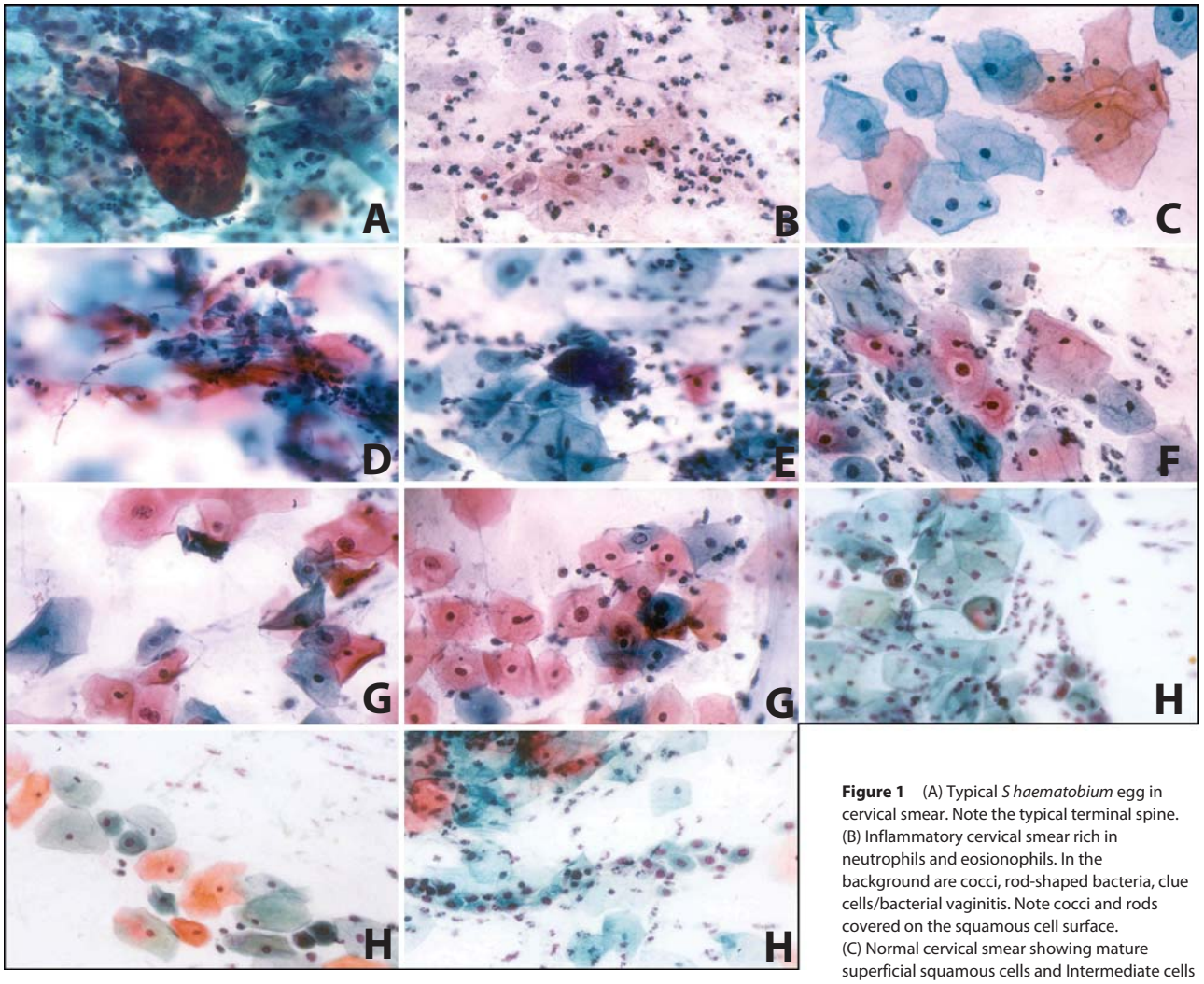

Figure 1 (A) Typical *S haematobium* egg in cervical smear. Note the typical terminal spine. (B) Inflammatory cervical smear rich in neutrophils and eosionophils. In the background are cocci, rod-shaped bacteria, clue cells/bacterial vaginitis. Note cocci and rods covered on the squamous cell surface. (C) Normal cervical smear showing mature superficial squamous cells and Intermediate cells with a clean background. (D) Moniliasis (*Candida albicans*) and pseudohyphae in cervical smear. (E) *Actinomyces*-like organisms. Note the cluster of nonbranching hyphae in cervical smear. (F) Koilocytosis in superficial squamous cells and intermediate cells. Note the irregular clearing of cytoplasm around the nucleus. (G) Mildly enlarged hyperchromatic nuclei and koilocytosis in superficial squamous cells and intermediate cells showing atypical squamous cells of undetermined significance. (H) Dyskaryotic superficial squamous and intermediate cells. Note the enlarged, hyperchromatic nuclei in squamous cells with koilocytosis, suggestive of squamous intraepithelial lesion (low/high grade) (A–H, Papanicolaou stain, ×450).

Table I *Pre– and Post–Papanicolaou Smear Findings in a* S haematobium*–Infected Woman*

Total duration IUD use (yr)	Change of device and duration of use	Pap smear findings
1–4	Copper T 200, 7 mo*	No pap smear done
5	Changed, reinsertion	Inflammatory smear with bacterial vaginitis
6 and 7	Copper T 200, 4 yr	Negative–normal smear
8	Changed, reinsertion	Clue cells/negative–normal smear
9	Copper T 200, 5 yr, 2 mo	Moniliasis
10	Changed, reinsertion	*S haematobium* egg
11	Copper T 200, 3 yr, 4 mo	*Actinomyces*-like organism
12	Changed, reinsertion	Koilocytosis, HPV with low grade ASCUS
13 and 1 mo	Copper T 380 in use	HPV-associated SIL, low/high grad

*Changed early for a medical reason (bleeding).

single dose of azithromycin. Barrier contraception was advised to prevent transmission of infection. At posttreatment follow-up, the patient was treated with metrogyl gel (local application).

The patient was followed up 2 years later, and her Pap smear revealed inflammation and ova of *S haematobium*. She could not be treated as she was not in the city. However, when she returned a year later she gave no history of any gynecologic complaints, and her Pap smear did not reveal any major cytomorphologic changes associated with schistosomiasis, as has been reported in the literature,[6,7] such as hyperkeratosis, inflammatory cellular changes, histocytes, increased number of polymorphonuclear leukocyes, erythrocytes or cellular atypia. Typical cytomorphologic diagnostic characteristics of *Schistosoma* are not frequent, and Pap smear screening for the detection of parasitic infections is well reported.[8-10] Retrospective analysis of yearly follow-ups with Pap smears for the past 13 years showed mild inflammatory changes associated with bacterial vaginosis (Figure 1B) followed by negative normal smears (Figure 1C), and then the smear revealed monilial infection (Figure 1D), followed by an *S haematobium* egg (Figure 1A) and followed by *Actinomyces*-like organisms (Figure 1E), which are generally associated with Cu IUD users.[11] Later, the smears revealed HPV infection associated with atypical squamous cells of undetermined significance (ASCUS) (Figure 1F). However, the annual follow-up Pap smear cytomorphology showed inflammation followed by HPV infection with low grade squamous intraepithelial lesion (Figure 1G). The yearly repeat Pap smear was done later and was again insignificant for the *S haematobium* egg, but cytomorphology revealed low grade squamous intraepithelial lesion (SIL) (Figure 1H).

The above findings of yearly follow-up of Pap smears showed that for the first 7 years Pap smears were negative for premalignant or malignant cells but mild inflammation associated with infections like bacterial vaginitis and moniliasis was observed. The *S haematobium* egg was observed in Pap smears in the sixth year. The yearly follow-up Pap smears from sixth year onwards showed HPV infection with ASCUS followed by squamous intraepithelial lesion with low/high grade (Table I).

Annual follow-up of contraceptive users is done routinely at the family welfare clinic with abdominopelvic speculum examination along with the routine Pap smear,[12] which helped to elicit these findings.

Discussion

Definitive diagnosis can be made only by documenting *Schistosoma* eggs in feces or urine or in a biopsy specimen. Eggs of *S haematobium* measure 120–160× 52–70 µm, with a characteristic terminal spine. The shell is stained bright orange by the modified Papanicolaou method. In addition, immunodiagnosis for schistosomiasis by enzyme-linked immunosorbent assay (ELISA) based on the detection of antigen circulating in the serum and/or urine has been developed.[1,13,14] However, Brown[15] noted that the patients excreting *Schistosoma* ova in urine elicited positive ELISA titers, whereas patients who were previously positive but no longer passing viable eggs were negative for ELISA. This shows that ELISA is useful for the detection of active infection only.

Genital disease manifestations of *S haematobium* occur frequently in women and men from endemic areas. Community-based studies from various countries in sub-Saharan Africa (endemic for schistosomiasis) indicate that between 32% and 75% of women infected with *S haematobium* have infection-associated lesions in the lower reproductive tract. The prevalence of lesions in the upper reproductive tract could be lower, but precise figures are not known. Gynecologic symptoms and abnormal physical signs may occur, such as postcoital, intermenstrual and postmenopausal bleeding; vaginal discharge; dysmenorrhea, ectopic pregnancy; and infertility, but these may not be direct consequences of the schistosomiasis.[16]

Urinary schistomiasis is usually asymptomatic but at times presents as dysuria, frequency, and terminal or total hematuria. At times it manifests as granulomas, pseudoabscesses, urethral hyperplasia, polyp formation, hydronephrosis, retrograde infections and renal failure. Chronic disease may lead to obstructive uropathy, chronic bacteriuria, bladder carcinoma and bladder calcification.[17-19] None of these symptoms were present in our patient. This may be because *Schistosoma* can survive and produce eggs for many years in the human body. However, the pathologic process is slow; the infected person may stay free from serious symptoms for many years. Genital diseases occur in the presence of *Schistosoma* egg excretion in urine and even in the absence of any egg excretion. A negative urine sample therefore does not exclude the existence of pathology in the genital tract.

There is circumstantial evidence that genital schistosomiasis in women is a risk factor for the bidirectional transmission of the human immunodeficiency virus[20] and that schistosomiasis of the cervix, with or without HPV infection, predisposes to the development of cervical cancer. It is known that women with *Schistosoma* eggs detected in a cervical biopsy may not excrete the eggs in their urine, and hence in endemic areas, control programs aimed at morbidity reduction take into consideration this type of disease manifestation.

The association of cervical carcinoma and *S haematobium* in cervical carcinoma cases has been reported by several investigators.[7,21,22] Similarly, the role of HPV as a cofactor in the genesis of cervical carcinoma has also been reported.[23,24] The increased risk of HPV infection in *Schistosoma*-infected cases may be because of traumatizing of the genital epithelium due to the spine of the *Schistosoma* egg. In our patient, yearly follow-up of the Pap smear showed the following: initial smears negative, followed by *S haematobium* infection, followed by HPV infection and then followed by HPV-associated low/high grade SIL over a period of 13 years and 1 month (Table I). These findings showed that HPV needs some medium to infect to initiate abnormal proliferation of squamous epithelium of the cervix. In the present case HPV infection was detected on cytomorphologic criteria reported in the literature.[25]

Theodor Bilharz, a German pathologist, from whom the disease took its original name, bilharziasis,[26] discovered the parasite re-

sponsible for the disease in 1851. The first report was published on gynecologic bilharziasis in cytologic material by Youssef et al in 1962[7] followed by Berry[6] and in wet smears by Swart and Van der Merwe in 1987.[27] In our case *S hematobium* in cervical smears proved useful in detecting the infection in an asymptomatic woman. This is the only case among the 109,480 cervical and endocervical smears screened in the last 30 years at this institute.

Recently Martinez-Giron et al[28] highlighted the value of various types of exogenous components as contaminants in routine cytologic smears: "uncommon structures simulating helminth eggs." Exogenous contaminants, such fiber, algae, spores and pollen grains, on cytologic smear sometimes can create confusion with such diagnoses as fungal hyphae, ova, cysts and larvae.[29-31] These findings emphasize the fact that one must be aware of rare parasites while screening routine Pap smears.[8-10]

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