

Infectious papillomavirus in the vapor of warts treated with carbon dioxide laser or electrocoagulation: Detection and protection

William S. Sawchuk, MD,^a Paul J. Weber, MD,^b Douglas R. Lowy, MD,^a and Leonard M. Dzubow, MD^b *Bethesda, Maryland, and Philadelphia, Pennsylvania*

Papillomavirus DNA has been reported recently in the vapor (smoke plume) derived from warts treated with carbon dioxide laser; this raises concerns for operator safety. We therefore have studied a group of human and bovine warts to define further the potential risk of wart therapy and to test whether a surgical mask could reduce exposure. Half of each wart was treated with carbon dioxide laser and the other half with electrocoagulation. The vapor produced by each form of therapy was collected with a dry filter vacuum apparatus and analyzed for the presence of papillomavirus. Vapor from human plantar warts was analyzed for the presence of human papillomavirus DNA, because there is no infectivity assay for human papillomavirus. Of plantar warts treated, five of eight laser-derived vapors and four of seven electrocoagulation-derived vapors were positive for human papillomavirus DNA. Greater amounts of papillomavirus DNA were usually recovered in the laser vapor than in the electrocoagulation vapor from the same wart. Bioassay readily detected infectious bovine papillomavirus in the vapor from bovine warts treated with either modality; more virus was present in laser-derived material. A surgical mask was found capable of removing virtually all laser- or electrocoagulation-derived virus, strongly suggesting that such masks can protect operators from potential inhalation exposure to papillomavirus. (*J AM ACAD DERMATOL* 1989;21:41-9.)

The increased availability and popularity of laser therapy has prompted concern for possible papillomavirus exposure for persons who treat warts with this modality. Laser operators are anecdotally said to have verrucae develop in unusual sites, such as the anterior aspect of the nares.¹ Experimental support for this concern has been provided by a recent report documenting that intact papillomavirus DNA can be isolated from the vapor (smoke plume) of warts treated with carbon dioxide (CO₂) laser.² Treatment of genital lesions carries an additional theoretic problem, because certain human papillomavirus (HPV) types that preferentially infect the genital region and mucous mem-

branes have been found in a majority of cervical carcinomas and in a few oral and laryngeal malignancies.

Although the findings of the report by Garden et al.² are highly suggestive, whether papillomavirus material in the vapor contained potentially infectious virus particles was not determined, nor was the potential virus exposure from lasers compared with that of another treatment modality. We have studied the relative amounts of virus isolated from the vapor of human and bovine warts treated with laser and with electrocoagulation, a treatment modality that has been widely used for many years for warts. We have analyzed the vapor from a bovine wart to determine whether it contains infectious virus, because a sensitive *in vitro* infectivity assay is available for bovine papillomavirus (BPV),³ in contrast to HPV. We also have examined the possibility that a surgical mask might help protect operators against potential virus exposure.

From the Laboratory of Cellular Oncology,^a National Cancer Institute, and the Department of Dermatology,^b The University of Pennsylvania.

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Reprint requests: Douglas R. Lowy, MD, Laboratory of Cellular Oncology, Building 37, Room 1B-26, National Cancer Institute, Bethesda, MD 20892.

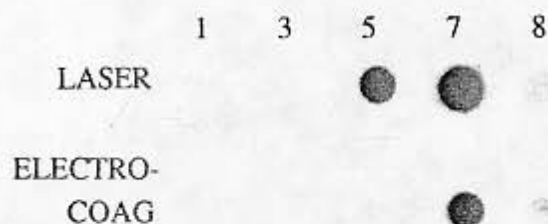


Fig. 1. Dot-blot analysis of DNA extracted from laser plume (upper set) and from electrocoagulation plume (lower set) of human plantar warts. Warts from subjects 1, 3, and 8 were hybridized with labeled HPV type 2 and subjects 5 and 7 with HPV type 1. Blots were washed under stringent conditions. Subject 3 is considered negative, subjects 1 and 8 are low positive, and subjects 5 and 7 are high positive.

MATERIAL AND METHODS

Treatment of warts

Eight persons with plantar warts that were morphologically flat and uniform were selected for study, because the amount of virus and viral DNA in plantar warts is usually large compared with that in other clinical types.^{4,8} All treatment was carried out in Philadelphia, Pa., by the same person (P. J. W.). The size of each wart was determined by measuring its surface area with the use of a grid composed of transparent 1 mm squares. Lesions were anesthetized with equal parts of 0.25% bupivacaine and 1% lidocaine without epinephrine (1 ml/cm² of tissue). Before therapy, a small shave biopsy specimen was taken (along the bisecting midline of the specimen, so that equal portions of each treatment half were included) from each wart for HPV typing. The specimen was placed immediately in lysis buffer (10 mmol/L TRIS-hydroxymethyl-amino methane, pH 8.0; 10 mmol/L sodium chloride; 10 mmol/L ethylenediamine tetraacetic acid; 0.5% sodium dodecylsulfate; and 50 µg/ml proteinase K) for storage and transport to Bethesda, Md., at room temperature.

Half of each wart was treated with a Xanar CO₂ laser, model No. XA-20 (Johnson & Johnson, New Brunswick, N.J.), with the use of a uniform 1 mm spot size at a setting of 10 W, a beam sweep speed of 3 mm/sec, and a peak intensity of 1270 W/cm², until the surface of the lesion and a 2 mm margin were completely charred. The char was vigorously removed with a saline-soaked sponge; then laser application and removal of char were repeated until there was no further bubbling of the wart tissue or until no excessive bleeding or friable dermis remained, which required an average of four to eight passes with the laser. The other half of each wart was treated by electrocoagulation, with the use of a Valleylab Surgistat (Boulder, Colo.). A 25-gauge needle applied to a Bernsco adapter (Bernsco

Table I. HPV DNA in vapor from human plantar warts

Patient	HPV type	HPV DNA in vapor material	
		Laser	Electrocoagulation
1	2	Low positive	Low positive
2	1	High positive	Not done*
3	2	Negative	Negative
4	2	Negative	Negative
5	1	High positive	Low positive
6	2	Negative	Negative
7	1	High positive	High positive
8	2	Low positive	Low positive

High positive, Visualized after 3-hour exposure; Low Positive, visualized after 12-hour exposure.

*The DNA in this sample was lost during extraction and could not be assayed.

Inc., Seattle, Wash.) was used as the uniform tip, which was inserted into a Valleylab disposable hand pencil unit, No. E2515. The coagulation level was maintained at a setting of 6, and passes were made over the wart at a rate of 2 mm/sec. The resulting char was removed with a curette; repeated electrocoagulation and curettage were then performed for an average of three to four cycles per lesion.

A previously excised bovine wart (kindly provided by Dr. Carl Olsen, University of Wisconsin, Madison), which had been stored at -70° C in Bethesda, Md., was hydrated for 12 hours in cold phosphate buffered saline, pH 7.4; frozen again at -70° C; and shipped on dry ice to Philadelphia. Thirty minutes before treatment with laser or electrocoagulation, the bovine wart was placed in sterile saline and allowed to thaw at room temperature. Equal volumes of wart, determined by fluid displacement, were treated with laser and electrocoagulation. Treatment of the bovine wart was carried out as for the human warts, except that the bovine wart was rotated with fine forceps during laser treatment, and curettage was performed with the wart held against a saline-soaked gauze pad during electrocoagulation.

Collection of vapor samples

The vapor from either procedure was collected with the use of a vacuum device (Stackhouse laser smoke evacuation unit [model No. LFA] with filter [model No. LFA-103]; Stackhouse Associates, El Segundo, Calif.). A 6-foot-long, corrugated, clear plastic hose (smallest internal diameter, 2 cm) was interposed between the collecting device and the LFA-103 filter. During collection of vapor from warts treated with either modality, the end of the collection device near the wart was maintained 2 cm from the wart surface. The

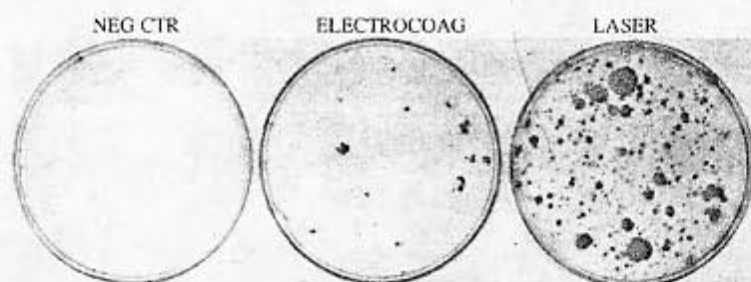


Fig. 2. Focal transformation induced in C127 mouse cells by vapor from laser or electrocoagulation treatment of a bovine wart. Dishes were stained 4 weeks after infection. Negative control (*NEG CTR*) dish of C127 cells shows no background foci. Dish with laser vapor-infected cells shows many foci. Dish with electrocoagulation vapor-infected cells shows fewer foci compared with that seen in laser dish.

vacuum unit was kept at a constant power level of 10, which uniformly generated a 42 L/min airflow (determined with a Timeter Instrument [Lancaster, Pa.] IIIB meter) without the interposed collecting device and a 40 L/min airflow with the device "in-line."

A special "dry" collecting device was constructed to trap airborne papillomavirus without passage through liquid. It was prepared to function by two mechanisms: direct gas/particle impact and Venturi effect. The device was a 15 cm clear plastic tube, with an internal diameter of 2 cm, to which a metal screw of 3 mm \times 7.5 cm had been affixed by metal wire to the exact center of the tube. A 1.6 cm metal washer was welded to a bolt and screwed to a fixed position along the screw. The washer was constructed to seat a piece of type HA filter paper, 1.9 cm diameter, 0.45 μ m pore size (Millipore Corp., Bedford, Mass.) with a 3 mm hole punched out of its exact center. Finally, three 1 mm screws were placed in the plastic tube approximately 4 mm proximal to the location of the washer. A 1.2 \times 7.0 cm strip of filter paper (grade No. 613, catalog No. 28310-128; VWR Scientific, Philadelphia, Pa.) was curled and inserted into the collection tube until it rested in the upwind position against the three screws. The unit was constructed so that gas and particulate matter would first reach the 1.9 cm filter disk. The airflow would increase behind the disk by the Venturi effect and deposit particulate matter on the nearby, now cylindrical, strip of VWR Scientific filter paper. Before each use, the air collection apparatus was cleaned three times with alcohol swabs. One VWR Scientific filter strip and one circular Millipore filter disk were used for each collection period; separate sterile instruments were used for each procedure. For testing the ability of a surgical mask to filter papillomavirus, a 4.4 cm disk cut from a mask (catalog No. 1818; 3M Surgical Products, St. Paul, Minn.) was placed over the collecting device, in front of the collection filter, and secured with a rubber band. After exposure to vapor, the mask and fil-

ter were analyzed separately for the presence of viral DNA.

Filters and masks to be analyzed for papillomavirus DNA were placed in vials with lysis buffer and shipped to Bethesda at room temperature. When treatment material was to be used in the infectivity assay, vapor was collected on a sterile VWR Scientific filter strip for 60 seconds, transferred to sterile tubes for transportation to Bethesda, Md., on dry ice, and stored at -70° C.

BPV infectivity assay

When used for infectivity studies, vapor-derived material collected on a filter was solubilized in 1.9 ml Dulbecco's modified Eagle's medium, with 50 U/ml penicillin and 50 μ g/ml streptomycin, and incubated 1 hour at room temperature, with occasional vortexing. The fluid was then pipetted into Eppendorf tubes (Brinkmann Instruments, Westbury, N.Y.) and centrifuged at 2000 revolutions per minute in an Eppendorf centrifuge (Brinkmann) for 5 minutes to remove the large debris.

The prepared extracts were assayed for infectious BPV by testing their ability to induce focal transformation of mouse C127 cells, as described.³ Briefly, 60 mm dishes seeded the previous day with 10^5 cells were infected with 0.5 ml of filter extract (obtained as described earlier) or 0.5 ml of a control BPV preparation (isolated directly from a bovine wart) known to contain about 100 focus-forming units of virus. Under the conditions used, infectious BPV particles will score as positive, but naked viral DNA will be negative. After 2 weeks' incubation, focal transformation of cells was readily detectable, and some dishes were fixed and stained (three parts 1% methylene blue, two parts methanol, and one part 1% carbol-fuchsin).

To confirm that the focal transformation observed was the result of infection with BPV, companion cultures were analyzed for the presence of BPV DNA by selecting for morphologically transformed cells and

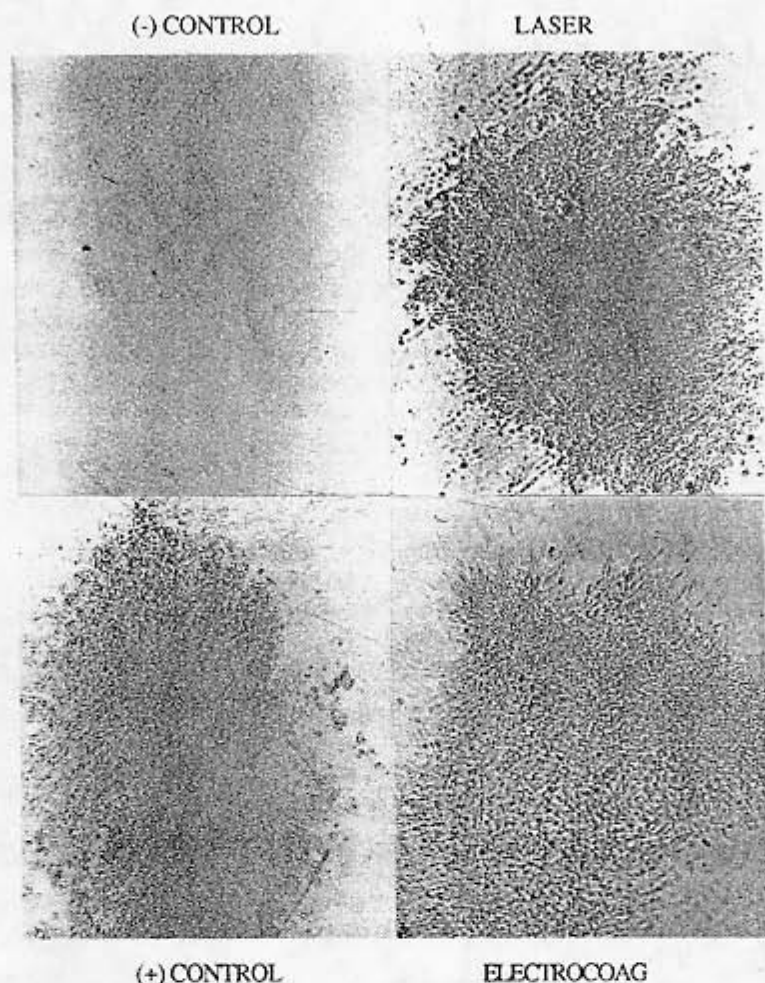


Fig. 3. Photomicrograph of individual foci induced by vapor from bovine wart. (-) *CONTROL*, C127 cells alone; (+) *control*, cells infected with known BPV extract; *LASER*, cells infected with extract from laser vapor; *ELECTROCOAG*, cells infected with extract from electrocoagulation vapor. (Magnification $\times 8$, except for *ELECTROCOAG*, which was $\times 12$.)

harvesting the cells for high-molecular-weight DNA extraction⁹ and Southern blot analysis.¹⁰ For each sample, 8 μg of high-molecular-weight DNA was digested with 25 units of *Cla*I restriction endonuclease under conditions recommended by the supplier (New England Biolabs, Beverly, Mass.), run on an 0.8% agarose gel to separate the DNA fragments, and transferred by capillary action to a nylon filter (Schleicher & Schuell, Keene, N.H.). The filter was baked, prehybridized, and hybridized according to the manufacturer's directions (Nytran product information; Schleicher & Schuell) with the use of cloned ³²P-labeled BPV-1 DNA as a probe. The BPV-1 DNA had been nick translated¹¹ with ³²P-dCTP (2000-3000 Ci/mmol; Amersham, Arlington Heights, Ill.), and 20×10^6 cpm (specific activity, 2.2×10^7 cpm/ μg) were used in each hybridization. After hybridization at 42° C overnight, the blot was

washed according to the manufacturer's directions, with a final stringent wash in 0.1 \times standard saline citrate (0.15 mol/L sodium chloride, 0.015 mol/L sodium citrate), 1% sodium dodecylsulfate at 65° C. The blot was placed in a photographic cassette with Kodak XAR 5 film and stored at -70° C until the film was developed.

Dot-blot analysis

To extract DNA from filters and masks on which vapor had been collected, the lysis buffer in each vial was increased to 3 ml and incubated at 37° C for 6 to 12 hours, with occasional vortexing. The solution was then deproteinized with phenol and chloroform-isoamyl alcohol, ethanol precipitated, washed sequentially with 70% and 95% ethanol, vacuum dried, and resuspended in 80 μl of 10 mmol/L TRIS-hydroxymethyl-amino methane,

pH 8.0, and 1 mmol/L ethylenediamine tetraacetic acid (TE). Thirty microliters of each resuspended DNA was mixed with 20 μ l of TE and 50 μ l of 20 \times standard saline citrate, denatured by boiling for 10 minutes, and blotted onto BA 83 nitrocellulose paper (Schleicher & Schuell) by use of a dot-blot apparatus. One microgram of high-molecular-weight human DNA from a person without warts was used as a negative control.

Before hybridization of the material described above, the small pretreatment biopsy specimens from each wart were used to determine the HPV type in the wart. In this procedure cloned HPV DNAs were blotted onto nitrocellulose paper with a dot-blot apparatus, and DNA extracted from each wart biopsy specimen was nick translated as described earlier and used as a probe to hybridize against the known HPV types. Blots were then washed under nonstringent (4 \times 20 minutes in 5 \times standard saline citrate, 0.1% sodium dodecylsulfate at 65 $^{\circ}$ C) or stringent (4 \times 20 minutes in 0.1 \times standard saline citrate, 0.1% sodium dodecylsulfate at 65 $^{\circ}$ C) conditions and exposed as described earlier. This analysis verified that the lesion contained papillomavirus DNA and determined the HPV DNA type in the wart. The DNA of the corresponding cloned HPV type was then used as the probe, after it was labeled by nick translation, to detect the minimal amounts of viral DNA expected to have been isolated from the vapor-derived material. After hybridization, the blots were washed under stringent conditions and processed for autoradiography.

RESULTS

HPV DNA in vapor from treated human warts

HPV type 1 was found in three warts; the other five had HPV type 2. HPV DNA was detected in five of eight laser-derived vapors and in four of seven electrocoagulation-derived vapors (Table I and Fig. 1). (The electrocoagulation vapor from one patient could not be analyzed.) There was a complete correlation between the two treatment modalities and the detection of viral DNA in the vapors. For either form of therapy, the vapors derived from warts with HPV type 1 tended to have significantly larger amounts of viral DNA than those derived from warts with HPV type 2. The vapors from all three warts with HPV type 1 (from subjects 2, 5, and 7) were strongly positive for viral DNA. In contrast, the vapors from only two of the five warts with HPV type 2 (from subjects 1 and 8) had detectable levels of viral DNA.

Vapor contains infectious papillomavirus

The results obtained herein indicated that viral DNA was present in the vapors derived from

treatment with laser or electrocoagulation, but they did not determine whether the papillomavirus material in the vapor was infectious. The current lack of a reproducible *in vitro* infectivity assay for HPV particles makes this assessment virtually impossible to perform for HPV. This problem can be circumvented, however, if bovine warts are used, because a sensitive, quantitative cell transformation assay is available for BPV.³ Portions of a bovine wart therefore were treated with laser or electrocoagulation, with the use of the same machines and settings and vapor collection system as for the human warts. The material derived from the bovine wart vapor was then tested for the presence of infectious BPV by assaying its ability to induce morphologic transformation of mouse C127 cells. Foci of morphologically transformed cells were found in material derived from both forms of therapy; the products of the laser vapor induced significantly more foci than those from electrocoagulation (Figs. 2 and 3).

To verify that the focal transformation had been induced by BPV, high-molecular-weight DNA was extracted from the morphologically transformed cells induced by the laser-derived extract and analyzed for the presence of BPV-1 DNA by the Southern blot technique, with the use of molecularly cloned BPV-1 DNA as the detection probe. The transformed cells contained BPV-1 DNA; *Cla*I digestion of the test DNA yielded the three expected BPV DNA fragments of 5.9, 1.3, and 0.6 Kb that hybridized with the BPV DNA probe and comigrated with those of the positive BPV-1 DNA control (Fig. 4). The results indicate that infectious BPV is present in the vapor-derived material.

Surgical mask blocks passage of virus

Having demonstrated that the vapor from both forms of treatment contains infectious BPV, we then tested whether placing a surgical mask in the vapor path could inhibit passage of the virus onto the collection filter. Vapor products derived from laser or electrocoagulation treatment of the bovine wart were collected on a filter, as previously described. For each form of treatment there were two 15-second treatment periods, first without the mask and then with the mask in place. The filters from each treatment period were analyzed by dot-blot hybridization for the presence of BPV DNA (Fig. 5). As expected, viral DNA was present on the filters used to collect the electrocoa-

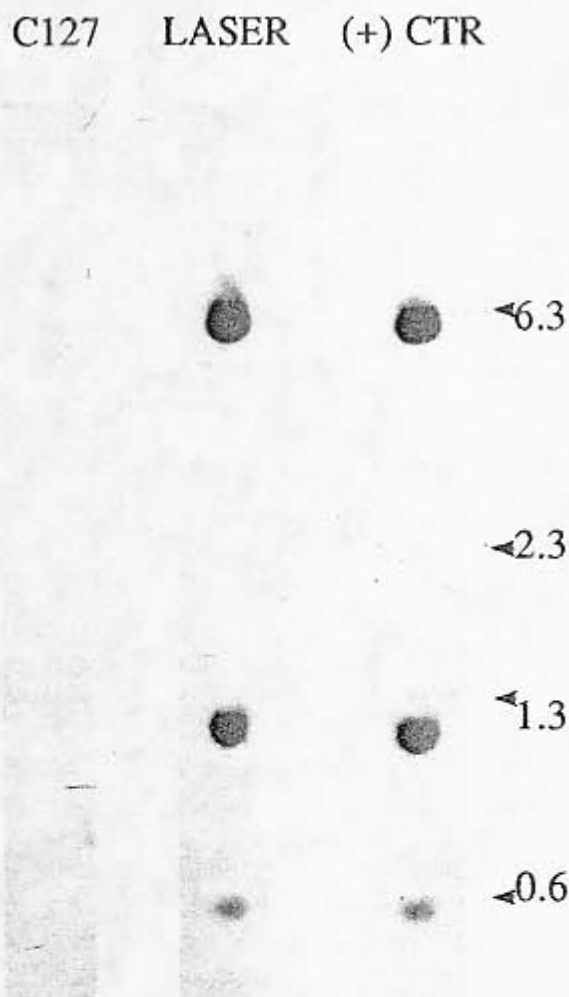


Fig. 4. Southern blot analysis of DNA from C127 mouse cells morphologically transformed by laser vapor. High-molecular-weight DNA was digested with *Cla*I. Blot was probed with labeled BPV-1 DNA. Negative control (C127 cells alone) shows no BPV DNA present. Positive control ((+)/CTR), C127 cells infected with known BPV extract and C127 cells infected with laser vapor products (LASER), show identical expected BPV DNA fragments (5.9, 1.3, and 0.6 Kb).

gulation and laser vapors produced before the mask was put in place. With either form of treatment, placing the surgical mask in front of the filter prevented the virus in the vapor from collecting on the filter. DNA was extracted from the masks to verify that the mask had indeed trapped the virus material and was found to contain substantial amounts of BPV DNA (Fig. 5). This result also confirmed that viral material continued to be present in the vapor during the treatment period with the mask in place and that airflow had been maintained during this time.

DISCUSSION

Laser smoke as an entity may represent a theoretic health hazard, because in rats exposed to long-term inhalation of CO₂ laser smoke, emphysema, bronchiolitis and congestive interstitial pneumonia developed.¹² In addition to the possible adverse effects of the smoke alone, it is important to know whether viable cells or microbial agents survive in the laser plume. Contradictory results have been reported. One group noted that S-91 melanoma cells appeared to survive in the neodymium laser plume,¹³ whereas other researchers showed a lack of tumor cell viability of the same S-91 melanoma line in the CO₂ laser plume.¹⁴ Efforts to detect viable cells in warts treated with CO₂ laser were reported to be unsuccessful.¹⁵ In one study¹⁶ bacteria were shown to survive CO₂ laser treatment, whereas in another study¹⁷ virtual destruction of the bacteria was demonstrated. As noted in the introduction to this article, intact papillomavirus DNA has been demonstrated in the laser plume from bovine and human warts treated with the CO₂ laser.²

Results of our study have confirmed and extended those in the report by Garden et al.² in several ways. In addition to detecting papillomavirus DNA in the laser plume from treated human and bovine warts, we also have documented, with a bioassay, that the plume from bovine warts contains infectious bovine papillomavirus particles (virions). We also have shown that papillomavirus, in somewhat lower concentrations, can be isolated from the plume resulting from treatment of bovine and human warts by electrocoagulation. Although our studies of infectious virus were limited to BPV, it is reasonable to infer that viable HPV virions may also persist in the plume derived from human material. Of perhaps greatest interest, we have shown that papillomavirus material in the plume can be efficiently trapped by a simple surgical mask.

We believed it was important to document that the plume contained infectious papillomavirus particles. Although naked papillomavirus DNA can be infectious,¹⁸⁻²⁰ the specific infectivity of virions is generally several orders of magnitude higher than that for naked DNA, so virions represent a much greater theoretic hazard than naked viral DNA. For example, viral DNA from the closely related polyomavirus (which is a member of the papovavirus group, as are papillomaviruses) is four to five orders of magnitude less infectious than virus

particles when both are administered parenterally to susceptible mice.²¹ We therefore performed the infectivity assay under conditions that would detect infectious virions but not potentially infectious viral DNA. The positive results obtained with this assay indicate that a significant proportion of the viral DNA detected in the plume was present in virions.

For our analysis of human warts, we studied plantar warts with HPV type 1 or HPV type 2. We monitored the presence of HPV material in the plume by assaying the viral DNA in the plume. In general we found more viral DNA in the plume from warts with HPV type 1 than from warts with HPV type 2. This result correlates with the observation that of all clinical wart types plantar warts containing HPV type 1 appear to be the richest in viral particles.^{4,8}

These results, in conjunction with what is known about the biologic characteristics of various types of papillomaviruses,²²⁻²⁵ may have some implications for the theoretic operator risk from the plume. (It is not likely that the plume material represents a significant hazard to the patient for distant spread, because the patient is presumably exposed chronically to his or her own virus.) Operator risk is principally to exposed areas, which means that the face, including the upper airway, and hands represent the major regions of potential concern. The presence of infectious papillomavirus virions in the plume may appear initially to represent a significant theoretic hazard. We, however, believe the actual degree of risk is not very great and that it can be reduced further by relatively simple precautions. It should be remembered that warts are not highly contagious, adults are not likely to develop nongenital warts, and genital warts are usually transmitted sexually. Although many health care workers apparently examine warts without wearing gloves, the incidence of hand warts among this group does not appear to be unusually high.

The clinical implications of the positive HPV results obtained in the vapor from electrocoagulation may be subject to various interpretations. Compared with the laser vapor, the amount of virus recovered was significantly less. We do not believe that the vapor from electrocoagulation represents a major risk. Physicians do not appear to be especially concerned about the risk of warts developing from this modality, and the lack of reports implicating electrocoagulation as a risk

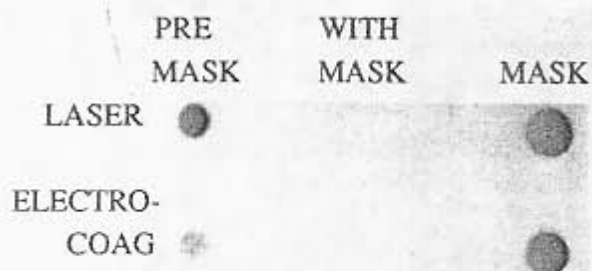


Fig. 5. Dot-blot analysis of BPV DNA from laser and electrocoagulation vapor blocked by surgical mask. Blots were probed with labeled BPV-1 DNA and washed under stringent conditions. *First column*, DNA extracted from filters exposed to vapor before placement of mask; detectable viral DNA. *Second column*, DNA extracted from filters exposed to vapor with mask in place; no detectable viral DNA. *Third column*, DNA extracted from masks used during exposure of filters in *second column*; abundant viral DNA was trapped.

factor may indicate that HPV exposure associated with this procedure may not be particularly hazardous. Electrosurgical devices are usually used in combination with curettage for treatment of warts, rather than continuously, as with the laser. Without the continuous vaporization and repeat passes expected with laser treatment, electrocoagulation treatment of warts results in less tissue destruction by the current. Laser plumes are dense, whereas the plume from electrocoagulation is much more diffuse. Our collection device was placed immediately (2 cm) above the treatment site; the operator's face is usually not located there during the procedure. The larger amounts of virus recovered from the laser vapor suggests that laser treatment may pose a greater theoretic hazard than electrocoagulation.

HPV types differ regarding concentration of virions that are typically present in lesions, their infectivity for different regions of the body, and their malignant potential. Because HPV type 1 lesions usually have the highest concentration of virus particles,^{4,8} lesions associated with this virus type would appear to pose the greatest theoretic risk for infection. This potential hazard is balanced by the benign nature of lesions induced by this HPV type. In addition, infection by HPV type 1 is usually restricted to the extremities. Therefore its theoretic risk to the face should be low, and wearing gloves (and shoes) should drastically reduce biologically important exposure. HPV type 2 lesions generally have a lower concentration of

virions. They therefore pose a lower theoretic hazard. Although HPV type 2 lesions are usually found on the extremities, they do appear to have a low level of infectivity for the face and the mouth.

Because vapor may be inhaled, the development of HPV infection of the upper airway mucosa represents an important theoretic concern for laser operators. The major risk of such lesions arising would be in association with treatment of genital warts, because the HPV types that are most frequently found in genital warts (typically HPV type 6 and HPV type 11)^{26,27} are also found commonly in patients with respiratory papillomatosis. The latter condition may affect any portion of the respiratory tree, from the external nares to the lung parenchyma, with a particular predilection for the larynx, and may be extremely difficult to treat.²⁸⁻³¹ In contrast to warts on the extremities, genital warts typically have low numbers of virus particles, although they may contain significant amounts of naked (nonvirion) viral DNA.³²⁻³⁵ Although almost all the viral DNA in HPV type 1-associated lesions (from the extremities) is found in virions, results of one study of HPV in condylomata showed that only 0% to 3% of the HPV DNA was in virions.³⁶ In another report of 18 condylomata that contained detectable levels of HPV DNA, no intact virions could be identified.³⁵ The paucity of virions in most genital lesions reduces their theoretic hazard. The HPV types most frequently associated with genital malignancies, such as HPV type 16, have been found rarely in tumors of the respiratory tract.^{37,38} The genital lesions containing these HPV types, however, usually contain very few virions.

Our results strongly suggest that the potential risk from inhalation may be markedly reduced by wearing a surgical mask, because in repeat experiments virtually all papillomavirus material was trapped by the mask. With the usual testing procedures,³⁹ the masks we used for the study are 99.7% effective in retaining particles 0.5 to 5 μm . Papillomavirus virions are only 55 nm in diameter. Because some particles in the laser plume are believed to be smaller than 0.5 μm , they might not ordinarily be trapped by the weave size of the mask alone.¹ Our results may suggest either that the particles in the laser plume that contain the virus or viral DNA are larger than the mesh size or that something else is trapping the smaller particles. In

favor of the latter explanation, the masks used are constructed with three webs, with a middle web composed of a polypropylene blown microfiber that is charged with a high, permanently embedded static charge (3M Company, personal communication). The charged microfiber middle web serves as an electrostatic filter which, in addition to the web of the mask, may help to trap smaller particles. For maximal protection, it presumably would be important for the mask to fit well, so that airflow around the edges is minimized. The ability of the mask to block passage of HPV is probably also relevant to preventing exposure to other infectious agents, including human immunodeficiency virus, that might be present in vapor.

We believe that the risk of papillomavirus infection for laser operators and other personnel can probably be minimal when proper precautions are taken. The most important precaution is proper and diligent use of a smoke evacuation system; when used appropriately with the orifice 1 cm from the treatment site, it is believed to be 98.6% efficient.⁴⁰ This efficiency, however, rapidly falls to 50% when the evacuator orifice is moved to just 2 cm from the treatment site.⁴¹ In addition, diminished amounts of papillomavirus in genital warts potentially represent a lower risk of the operator, considering the exposure to fewer viral particles along with the relative tissue specificity of HPV. Finally, the use of a properly fitted and tied surgical mask similar to the one we tested should greatly reduce the risk of airway exposure to virus.

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