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Laboratory-acquired vaccinia infection

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Abstract

Background: Complications following vaccination with vaccinia virus have been well described but are not commonly observed. The use of vaccinia as a tool in molecular biology, in the development of therapeutics, and the anticipated increase of vaccinations in the general population due to the threat of bioterrorism have created a renewed awareness of the post-vaccination complications and the consequent need for clinical and laboratory diagnosis. *Objectives:* To report the clinical presentation and subsequent diagnosis of generalized vaccinia that resulted from a laboratory accident in an unvaccinated subject. *Study design:* The patient was seen by a local infectious disease's specialist and evaluated clinically and with laboratory support relative to a differential diagnosis. *Results:* Careful assessment of the patient's history, an evaluation of the workplace, and the elimination of likely microbial etiologies led to the diagnosis of generalized vaccinia. Laboratory confirmation was obtained by use of electron microscopy (EM) to observe poxvirus particles in infected cell cultures. *Conclusions:* Exposure to vaccinia virus should raise the index of suspicion for patients with skin lesions. Rapid diagnosis may be accomplished by direct examination of lesion material by EM. The virus also readily replicates in commonly available cell cultures and in the absence of immune reagents, typical poxvirus particles may be observed in the infected cells by EM.

Keywords: Vaccinia virus; Laboratory acquired infection; Generalized vaccinia

1. Introduction

The global eradication of smallpox in nature eliminated the need for population-based vaccination that utilized vaccinia virus. Subsequent to eradication, vaccinia became a tool in recombinant research (Binns and Smith, 1992) and a potential vector for therapeutic applications (Paoletti, 1996). The Advisory Committee on Immunization Practices (ACIP) has recommended vaccination of laboratory workers who are at risk of vaccinia infection (CDC, 2001). However, complications associated with vaccination are well documented (Lane et al., 1969). Recent events have generated considerations of smallpox as a weapon of bioterrorism (Henderson, et al., 1999) and have prompted public preparations that include vaccination (LeDuc and Jahrling, 2001). Consequently, resumption of vaccination will result in increased numbers of vaccinees showing side effects and the need for accurate clinical assessment and appropriate treatment. To that end we report a case of laboratory-acquired infection with vaccinia and discuss the clinical presentation, diagnosis, and management.

2. Case report

A previously well 25-year-old research technician who had not been previously vaccinated was working with vaccinia virus and accidentally cut her left second finger on a cover slip. Twelve days later a lesion, which she described as a pimple, developed at the site of the cut. She squeezed the pimple and a drop of pus squirted onto her face. She first noted a lesion on her chin a day or two later. Over the next several days both lesions grew progressively worse; auxiliary and submental adenopathy were first noted at day 14, and she developed generalized malaise and a fever to 102.9 at day 17. Physical examination on day 18 showed a 1-cm violaceous, umbilicated bulla with

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marked surrounding inflammation on the palmar surface of her left second finger, and over the distal interphanlangeal joint (Fig. 1A); two similar but smaller lesions were in the submental area (Fig. 1B). On day 20 four additional small vesicles developed, one on each plantar surface, one in the left poplitial fossa, and one over the right scapula. On the same day, she defervesced and felt systemically better.

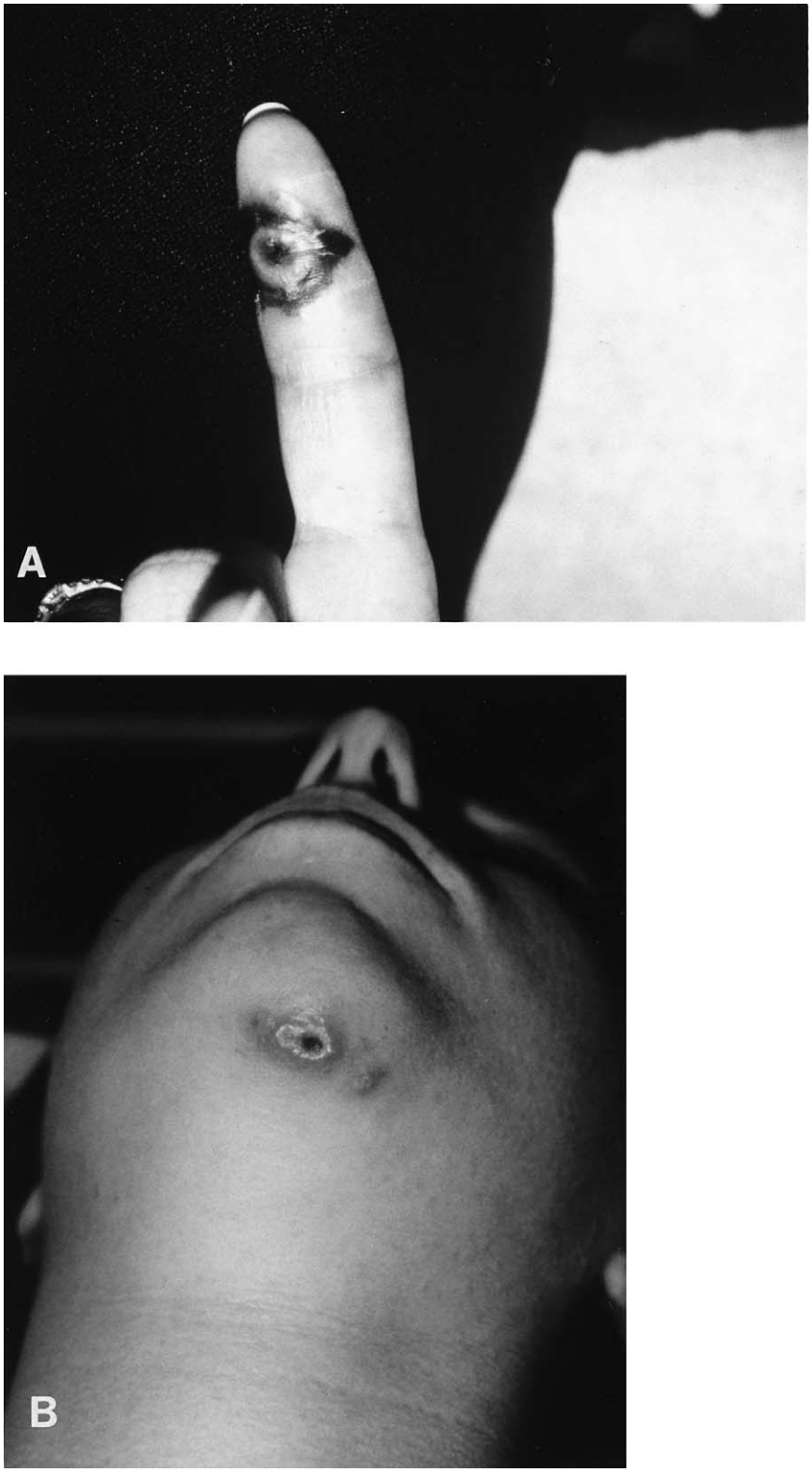

Fig. 1. Vaccinia finger (A) and chin (B) lesions.

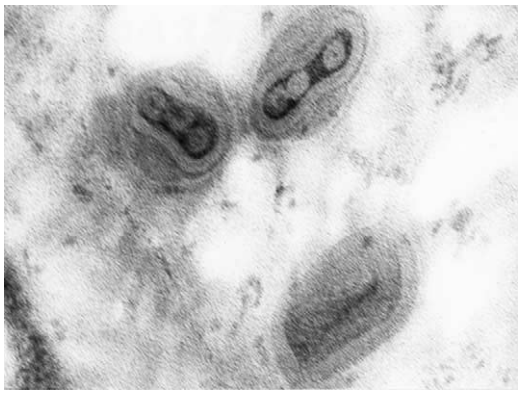

Fig. 2. Poxvirus virions seen in infected primary rhesus monkey kidney cells at 150 000×.

The patient was initially treated empirically with trimethoprim/sulfamethoxazole and then cephalexin. Vaccinia immune globulin, ribavirin and cidofovir were not given. On day 28 the skin lesions were fading although she continued to complain of easy fatigability. On day 36 the finger lesion had decreased to a 3-mm blackened eschar and she felt almost back to full strength.

Five days after the initial appearance of the finger blister, an aspirate of the blister was taken for bacterial, fungal, and viral cultures. Bacterial and fungal cultures were negative. The aspirate was inoculated onto monolayers of primary rhesus monkey kidney (PRMK) and MRC-5 cell cultures. A cytopathic effect (CPE) was seen in PRMK and MRC-5 cultures at 2 and 3 days, respectively, after inoculation. The CPE was extensive and was not typical or recognizable of viral agents commonly isolated. Immunofluorescent tests performed on infected cells with monoclonal antibodies to herpes simplex virus and varicella zoster virus were negative. Negative-stain electron microscopy (EM) performed on culture fluids from infected cell cultures was negative. Subsequently, infected cell cultures were centrifuged, the cell pellet fixed in gluteraldehyde and embedded in Epon 812; the resulting block was sectioned and counterstained. Particles with typical poxvirus morphology were readily observed (Fig. 2). Neutralizing antibody to vaccinia was not detectable in serum obtained from the patient 2 weeks after infection but was detected 25 days after infection. The only other test performed was a CBC and it was normal.

3. Discussion

Infection likely occurred through contamination of the finger laceration and was probably transferred secondarily to the chin through autoinoculation. When vaccinia is used as a vaccine, the consequences of infection may be observed at the site of inoculation in 3–4 days. Since the interval between the cut and infection was 12 days in our case, it is likely that the cut was inoculated some days after it occurred. Indeed, she continued to work with vaccinia over the ensuing days. The four additional vesicles that developed fit the description of generalized vaccinia, a nonspecific term applied to the vesicular rash that is blood-borne and may appear after vaccination (Neff, 2000). Generalized vaccinia should be distinguished from progressive vaccinia that can occur in immunocompromised hosts where it maybe fatal and from the spread seen in atopic skin lesions, known clinically as eczema vaccinatum.

The clinical and laboratory diagnosis, and the management of vaccinia infections have become an anachronism, unfamiliar to new generations of virologists and clinicians. Yet, vaccinia's new roles in recombinant research, as a potential vector for therapeutics, and the bioterror-driven revival of vaccination against smallpox, have reintroduced concern about vaccinia's potential complications. Management is supportive for local lesions and mild generalized vaccinia, as attested to by the approach used in this case. Progressive vaccinia has been reported to have been treated successfully with a combination of ribavirin and vaccinia immune globulin (Kesson et al., 1997). It should be noted that surgical manipulation could promote local spread (Klingebiel et al., 1988) and treatment with steroids could promote systemic dissemination (Casemore et al., 1987). Marked inflammation may suggest bacterial infection and inspire antibiotics as occurred initially in this case.

Close consideration should be given to the clinical presentation and the local epidemiology when considering laboratory diagnosis. In this case, a conventional protocol for identifying an unknown viral agent was followed. Initially, vaccinia was not high on the index of suspicion but with the failure of bacterial and fungal cultures to isolate an agent and the failure to make an identification with available viral reagents, EM was applied to cell cultures showing CPE. When vaccinia is suspected, however, negative stain EM is a rapid and sensitive procedure when utilized with lesion specimens (Nakano and Esposito, 1989) and should be considered as a first approach. In the absence of EM, infected cell cultures may be stained by immunofluorescence (IF). Antibody to vaccinia virus is available commercially and can be utilized in an indirect IF format. The test is likely to only give confirmation of an orthopoxvirus and not vaccinia since most available reagents cross-react with all orthopoxvirus species (personnel communication).

Enforcement and monitoring of safety measures utilized in the laboratory is essential. Following this incident, procedures for handling contaminated glassware were reviewed and changed to prevent a repetition of this incident. Importantly, employees should also be educated about the potential hazards of their work environment, including autoinoculation and the potential spread to contacts. The advantages and disadvantages of vaccination for laboratory personnel have been discussed (Buller and Palumbo, 1992; Williams and Cooper, 1993; Wenzel and Nettleman, 1989; Perry, 1992). The ACIP recommends that personnel working with vaccinia should be vaccinated (CDC, 2001).

With the recent impetus to resume smallpox vaccinations out of concerns over bioterrorism, accidental exposures and injuries (e.g. needle sticks) and syndromes similar to this case may become more common.

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