SHORT ARTICLES

VIRUS TRANSMISSION BY SUBCUTANEOUS JET INJECTION

P. R. G. BRINK, A. M. VAN LOON,* J. C. M. TROMMELEN, F. W. J. GRIBNAU,†
I. R. O. SMALE-NOVAKOVA‡

*Department of Surgery, Groot Ziekengasthuis, Nieuwstraat 34, 's-Hertogenbosch and *Department of Medical Microbiology and Divisions of ‡Haematology and †General Internal Medicine of the Department of Medicine, St Radboud Hospital, Nijmegen, The Netherlands*

SUMMARY. An animal model was used to establish the risk of transmitting a virus infection by subcutaneous jet injection. Virus transmission was studied with mice chronically infected with LDH virus. The virus infection was transmitted by subcutaneous jet injection in 16 cases out of 49. Other routes of cross-infection were ruled out. Before using the jet injector as a harmless instrument for mass subcutaneous injection, further experiments on the risks of virus transmission should be performed.

INTRODUCTION

Since the clinical introduction of the jet injector for subcutaneous and intracutaneous administration of fluid by Hingson and Hughes (1947), the method has been widely accepted. Although it is an invasive technique, and the same injector is used for many patients without intervening sterilisation, only a few investigators have examined the risks of transmitting virus infections. The absence of an explosion of hepatitis after the large-scale use of jet injection for subcutaneous administration of fluid does not necessarily exclude this possibility. Therefore, we studied in an experimental animal model the risk of transmitting a viral infection when using jet injection. Lactic dehydrogenase (LDH) virus was used in this study because this virus causes a persistent infection, accompanied by a viraemia with high titres of infectious virus in the blood of infected mice (Notkins, 1965). The virus causes a significant rise in the LDH activity of serum, which is independent of the infective dose. Therefore, infection may be detected easily by measuring the LDH activity in plasma *c.* 3 days after inoculation.

MATERIALS AND METHODS

Animals. Male Swiss ICR mice between 8 and 12 weeks old were used. The mice were caged separately, preventing mutual contact to exclude cross-contamination by faeces and urine. Blood was obtained from all mice by retro-orbital puncture with heparinised micropipettes. Plasma was separated from the cells by centrifugation at 12500 *g* for 4 min and was assayed directly. Haemolysed specimens were discarded.

Lactic dehydrogenase assay. LDH was assayed by the method of Wroblewsky and La Due with an Abbott bichromatic analyser.

Jet injection. A compressed air Med-E-Jet-injector (Med-E-Jet-Corp., Cleveland, USA) was used throughout this study. The smallest possible volume (0.05 ml) of sterile physiological saline was injected into the preshaved thighs of mice by holding the nozzle firmly against the skin. The fluid pressure of each shot was 280 bar, with a nozzle diameter of 0.15 mm.

RESULTS

At the beginning of the experiment, the LDH activity was determined in all mice (table) to exclude previous infection with LDH-virus. Next, the LDH activity was determined in a small

Received 15 Jan. 1985; accepted 26 Mar. 1985.

TABLE

Group	Number of mice	LDH activity (U/L; mean $\pm$ SD) in serum	
		before jet injection	after jet injection
Control	84	725 $\pm$ 285	—
A	6	540 $\pm$ 410	704 $\pm$ 285
B	6	670 $\pm$ 135	3785 $\pm$ 650
C	22 (23)	758 $\pm$ 185	3475 $\pm$ 945
D	33 (49)	720 $\pm$ 265	665 $\pm$ 175
	16	770 $\pm$ 265	3240 $\pm$ 260

For groups C and D, LDH activities are derived from numbers of mice in the first column; numbers in second column in parenthesis are totals in group. See text.

group of mice (A) 3 days after subcutaneous jet injection of 0.1 ml of saline. No significant increase of LDH activity was found. Similarly, the LDH activity was determined in a group of mice (B) which had been given, 3 days earlier, intraperitoneal injections of plasma containing 10^5 ID 50 of LDH virus/ml. Infection was confirmed by increased LDH activity after 3 days. To ascertain the transmissibility of the LDH virus, transmission of infection from this group of infected mice (B) to 23 mice (group C) by simple subcutaneous pricking with a 26 G needle, without injection of fluid, was attempted. A significant increase of LDH activity, indicating successful LDH virus transmission, was observed in 22 of the 23 mice. The whole population of infected mice was used for further investigation.

Under ether anaesthesia, an infected mouse, pre-shaved and cleaned with ether, was given 0.05 ml of saline by jet injection (fig. 1). Any wounds created during shaving, spilling of injection fluid or bleeding were noted. The nozzle was then cleaned with an ether-saturated cotton ball. One minute later 0.05 ml of saline was given to a susceptible mouse by jet injection from the same

FIG 1.—Jet injection procedure on the preshaved thigh of a mouse.

FIG. 2.—LDH activity in plasma from 49 mice of group D, before and after jet injection, using a possibly contaminated nozzle. Increased activity after 7 days in 16 mice indicated that they had become infected with LDH virus.

injection head. Afterwards, the skin was cleaned with ether. Spilling, wounds and bleeding were noted. Before the next shot, the nozzle was disinfected in boiling water for at least 2 min to kill the LDH virus. Forty-nine pairs of mice were treated in this way (Group D). Seven days later, the LDH-activity was determined in the plasma of the susceptible mice. Significantly increased LDH levels were found after 7 days in 16 of these 49 mice (32.7%), suggesting infection with LDH virus. Mean LDH activity in this group of 16 mice was 3240 ± 260 U/L (fig. 2). To exclude other causes for the rise of LDH levels in these 16 mice, plasma from each of them was injected intraperitoneally into 16 susceptible mice. A significant five-fold increase of LDH activity was found in all these latter mice, confirming LDH virus infections in the 16 mice of group D.

DISCUSSION

Evidence for the safety of jet injection, with respect to possible transmission of virus infections, is well documented in the literature and mostly based upon retrospective and

mechanical investigations. The absence of an epidemic of infectious hepatitis after vaccinating more than half a million people suggests that the method is safe (Barrett and Molner, 1962). However, most jet injections for vaccination are intracutaneous injections and require a special spacer surrounding the central nozzle to be applied firmly to the skin. The nozzle does not touch the skin, but closely approximates it (Lipson *et al.*, 1958). Subcutaneous injection requires a direct contact between nozzle and skin. After inoculation, a small amount of blood can frequently be seen at the puncture site some time after removing the head of the jet injector. Tests for occult blood on the nozzle after jet injection have been done. In 300 cases of intracutaneous injection one such test was positive. This occurred when an individual received a second dose of vaccine in the same site as the first (Lipson *et al.*, 1958). Darlow (1970), after precipitating the serum contaminating the nozzle with ^{131}I -labelled antibody to human serum, noted a small rise of radioactivity. The risk of contamination of the nozzle during intracutaneous injection appears minimal. Different kinds of valve mechanism should prevent a flow-back of injection fluid which might be contaminated (Spiess, 1975).

Multiple injections *in vitro* into tissue infiltrated with radioactive isotope did not cause increased radioactivity on the head of the jet injector (Black *et al.*, 1978). After jet injections into two patients with chronic hepatitis B, Abb *et al.* (1981) could not demonstrate HBV-associated antigens on the nozzle by radio-immune-assay techniques.

The investigations cited above were designed to exclude the risk of transmission of virus infection by intracutaneous jet injections. However, subcutaneous jet injections might have a higher risk of contamination, because of the direct contact between skin and nozzle. Mass jet injection has been used for subcutaneous injection of low doses of heparin, because this method is much cheaper and less time consuming than using a syringe and needle and not painful for the patient (Black *et al.*, 1978; Held *et al.*, 1981; Harenberg *et al.*, 1982). The rise of LDH activity in 16 mice after subcutaneous jet injection suggests a high risk of virus transmission. The increase of LDH activity could not be explained by tissue damage, in view of the slight increase of LDH activity after jet injection of saline in 6 mice (group A). We found that needle transmission of LDH virus was very frequent (95.6%, Group C), making this investigation highly sensitive. (Theoretically less than 10^{-7} ml of serum of the acutely infected animal could contain enough infective particles to transmit the infection (Notkins, 1965)).

Cross-infection between infected and non-infected mice was excluded by housing them in separate cages. Transmission by faeces, urine and feed was excluded. Thus, the jet injector was the only possible route of transmission. No relation was found between transmission and spilling, splashback or bleeding. Postinjection bleeding was relatively uncommon (4%). Spilling or splashback in some form, however, was seen after almost every shot. The volume of fluid injected into each mouse corresponded with an injection of approximately 175 μl of fluid, in less than 0.1 sec, in man. Probably the enormous tissue pressure caused a splashback of the injected fluid. This retrograde stream could be responsible for the transport of virus particles. It has been found that splashback or spilling is much less from human skin. Extrapolation from mouse to human is therefore difficult. Further experiments must be performed before we can say that mass injection by jet injector carries no risk of virus transmission. Until then it might be justifiable to screen for HBsAg when a patient belongs to a high risk group, or to abandon the jet and give the treatment with individual syringes and needles.

REFERENCES

- Abb J, Deinhardt F, Eisenburg J 1981 The risk of transmission of Hepatitis B virus using jet injection in inoculation. *Journal of Infectious Diseases* **144**: 179.
- Barrett C D, Molner J G 1962 Automated multiple immunization against diphtheria, tetanus and poliomyelitis. *Journal of School Health* **32**: 48-49.
- Black J, Nagle C J, Strachan C J L 1978 Prophylactic low-dose heparin by jet injection. *British Medical Journal* **2**: 95.
- Darlow H M 1970 Jet vaccination. *British Medical Journal* **4**: 554.
- Harenberg J, Arleth D, Köppern K, Matthes K, Zimmerman R 1982 Subkutane Applikation von Heparin mit einem Injektor. *Deutsche Medizinische Wochenschrift* **107**: 497-500.
- Held F, Meyer-Menk W, Hüter J 1982 Eine neue Technik zur Anwendung der "Low dose" Heparin-thrombose-Prophylaxe. *Intensivmedizinische Praxis* **5**: 39-44.

- Hingson R A, Hughes J G 1947 Clinical studies with jet injection: A new method of drug administration. *Current Researches in Anesthesia and Analgesia* **26**:221-230.
- Lipson M J, Carver D H, Elepp M G, Hingson R A, Robbins F C 1958 Antibody response to poliomyelitis vaccine administered by jet injection. *American Journal of Public Health* **48**: 599-603.
- Notkins A L 1965 Lactic Dehydrogenase Virus *Bacteriological Reviews* **29**:143-160.
- Spiess H 1975 Hepatitisübertragung durch Hochdruckinjektion? *Deutsche Medizinische Wochenschrift* **100**:2465.