

leads to implosion of the immune system. There is evidence that HIV may trigger an autoimmune condition that leads to destruction of the immune system. This is supported by our observation that the HLA haplotype A1, B8, DR3 is associated with a more rapid progression to AIDS.³ This haplotype is also associated with autoimmune disorders such as glomerulonephritis and systemic lupus erythematosus. Our results therefore lend credence to the autoimmune hypothesis, as do the observations of Dalgleish and colleagues, reviewed in your April 4 editorial. Again, nothing in our research suggests that a particular HLA type or any other characteristic of the immune system is an absolute requirement for progression to AIDS once HIV infection is established.

Whilst I support, and encourage the reporting of, minority views there is an equal obligation on the part of the media to record other results and opinions fairly. If the belief that AIDS is not due to HIV becomes prevalent, those so convinced will feel justified in disregarding precautions designed to prevent viral spread. *Lancet* readers might feel that this will be at the very least, unfortunate—and at worst could lead directly to the deaths of countless misinformed individuals.

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1. Carr R, Veitch SE, Edmond F, et al. Abnormalities of circulating lymphocyte subsets in haerophilias in an AIDS-free population. *Lancet* 1984; i: 1431-34.
2. Ludlam CA, Tucker J, Steel CM, et al. Human T lymphotropic virus type II (HTLV-II) infection in seronegative haerophilias after transfusion of factor VIII. *Lancet* 1985; i: 233-36.
3. Steel CM, Ludlam CA, Barton D, et al. HLA haplotype A1B8DR3 as a risk factor for HIV related disease. *Lancet* 1988; i: 1189-88.
4. Dalgleish AG, Wilson S, Gompels M, et al. T-cell receptor variable gene products and early HIV-1 infection. *Lancet* 1992; 339: 824-27.

Origin of AIDS

SIR.—The hypothesis that a poliovaccine used in central Africa in the late 1950s was contaminated by simian immunodeficiency virus (SIV), a possible precursor to HIV-1, has been disputed on three grounds: the inability of monkey kidney cells to support SIV/HIV infection; the implausibility that oral administration of vaccine could initiate viral infection; and that SIV could have evolved to HIV-1 in the two decades between administration of the poliovaccine and the emergence of HIV-1 in central Africa.

Monkey kidney was not the only type of tissue culture used for the manufacture of oral poliovirus. The Wistar Institute, Philadelphia, in the late 1950s and early 1960s developed human diploid lines from embryonic tissues for use in the production of poliovaccine and other vaccines from viral seed stocks. Fetal lung proved the most promising and these cells could withstand 50 or more serial cultivations. At that time, human poliovirus vaccines (attenuated and killed) were acceptable only when grown in primary monkey kidney, since such tissue was presumed to have no malignant properties.¹ For the purpose of poliovaccine production, a human diploid line was inoculated with the CHAT strain of type 1 poliovirus cultured and attenuated in monkey kidney, a source of simian viral and retroviral contamination. The poliovaccine was then frozen and shipped to the site where the mass vaccinations were to be held. WI-38 (human fetal lung cell) strain was reportedly used to prepare poliovaccine between 1961 and 1963 for vaccinations in the United States, Sweden, Switzerland, and Yugoslavia.² Culture of human diploid lung fibroblasts, on infection with HIV-1, will produce and release infectious virus.³ Between August, 1958, and April, 1960, over 75 000 children under five years of age were immunised in Léopoldville, Belgian Congo, with vaccines prepared at the Wistar Institute. Is there any possibility at all that those poliovaccines were developed in the human cell line? I can find no evidence for this but the possibility deserves an airing.

In the African trial, vaccine was diluted in neutral saline on the day of vaccination and placed in semi-automatic syringes. "An attempt was made to squirt the vaccine into the back of the child's throat so that swallowing was involuntary."⁴ Subclinical viral infection may be initiated by spraying of contaminated vaccines into the nasopharyngeal cavity if the vaccine enters the respiratory system. Morris et al used an inoculum containing SV40 and administered it via nebuliser into the nose and mouth, resulting in

respiratory infection.⁵ Alveolar macrophages and fibroblasts are susceptible to infection by HIV-1 and, probably, by SIV too. Retroviral infection of recipients of a nebulised contaminated vaccine remains a possibility.

Dr G Myers (an expert in gene sequencing and head of the Los Alamos National Laboratory) and colleagues⁶ ask in a 1992 article in an AIDS journal, whether, as a "starting point for inquiry", HIV might simply be SIV adapting to a human host. "There is no clear answer to this question at this time; however, the notion is less far-fetched in 1992 than it was merely a few years ago..." Furthermore lentiviruses "appear to have an enormous potential for diversification that is not witnessed in other retroviruses. As a consequence... ecological and evolutionary dynamics become tightly intertwined with the lentiviruses."⁶

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1. Haytick L, Moorhead P. The serial cultivation of human diploid cell strains. *Exp Cell Res* 1961; 25: 585-621.
2. Haytick L. A comparison of primary monkey kidney, heteroploid cell lines, and human diploid cell strains for human virus vaccine preparation. *Am Rev Respir Dis* 1963; 88 (suppl): 387-93.
3. Dolei A, Serra C, Arca M, Tonello A. Acute HIV-1 infection of CD4+ human lung fibroblasts. *AIDS* 1992; 6: 232-34.
4. Lebrun A, Cerf J, Gelfand H, Courtois G, Plotkin S, Koperovskii H. Vaccination with the CHAT strain of type 1 attenuated poliovirus in Léopoldville, Belgian Congo. *Bull WHO Org* 1960; 22: 203-13.
5. Morris J, Johnson K, Aulaisio C, Chanock R. Clinical and serologic responses in volunteers given vaccinating virus (SV40) by respiratory route. *Proc Soc Exp Biol Med* 1961; 106: 56-59.
6. Myers G, MacInnes K, Korber B. The emergence of simian human immunodeficiency viruses. *AIDS Res Hum Retrov* 1992; 8: 373-86.

HIV screening in Russia

SIR.—In the May 9 issue of *The Lancet*, Round the World correspondents discussed AIDS-associated problems in former Eastern bloc countries (Czechoslovakia and Poland). I would like to emphasise another alarming concern—namely, the rapid growth in false-positive HIV tests in the former USSR, and in Russia especially. In 1990, of 20.2 million HIV tests done in Russia only 112 were confirmed and about 20 000 were false positives. 1991 saw some 30 000 false positives out of 29.4 million tests, with only 66 confirmations.¹

Such huge numbers of false positives are predictable for mass screening for HIV in low prevalence populations, such screening being a major component of the AIDS control strategy in Russia. The severe consequences can largely be avoided if the HIV status of screening-test positives is clarified rapidly, but this does not happen in Russia. The time scale for completing confirmation tests ranges from weeks to months and field epidemiologists often embark on contact tracing without waiting for the confirmation. Also, personal information on results of HIV tests is often leaked, resulting in discrimination and stigmatisation both of HIV-infected individuals and the large number of false positives.

Changing attitudes in Russia to the non-discriminatory acceptance of HIV-infected people and sexual minorities will take time. A consensus among health professionals, administrators, and legislators on an AIDS control strategy has yet to be reached, but the social, emotional, and other difficulties associated with HIV false positivity in Russia could be lessened by reducing the number of mandatory HIV tests in groups where HIV prevalence is low. Pregnant women, for whom two tests during pregnancy are mandatory, could benefit the most of all because in 1991 alone some 8000 false-positive results were reported in pregnant women, with only 6 confirmations. A quarter of HIV tests in Russia are done in this group, so stopping or reducing mandatory HIV screening, at least in pregnant women, would make it possible to improve the financing of more essential AIDS-related services and projects.

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1. Voevodin AP, Naumov SA, Fomochov AE, Shutova AP, Sicheva TB. HIV screening in Russia: false positive problem. VIII International Conference on AIDS (Amsterdam, 1992); abstr 6265.

HIV-1 viraemia and influenza

SIR.—Cellular activation of CD4 lymphocytes is required for HIV-1 replication in vitro and it is often assumed that immune stimulation due to infection, vaccination, or allogeneic exposure can result in enhanced HIV-1 replication in vivo. I have examined this possibility by serial measurement of viral burden in blood from three HIV-1 seropositive, symptomless individuals who had acute influenza infection or influenza vaccination. Infectious titres of HIV-1 in plasma and peripheral blood mononuclear cells (PBMC), measured by end-point-dilution cultures,¹ increased transiently in patient 1 who had an acute illness clinically diagnosed as influenza. The levels of infectious HIV-1 in plasma increased by 20-fold in the two given influenza A vaccine:

Case	Day	HIV-1 titre	
		Plasma (TCID ₅₀ /ml)	PBMC (TCID ₅₀ /10 ⁶ cells)
1	-91	1	100
	-27	1	100
	4	100	1000
	14	5	100
	35	1	100
2	-22	5	10
	0	5	10
	10	100	100
3	0	25	1000
	7	500	1000
	22	25	1000
	22	25	1000

TCID₅₀ = tissue-culture infective doses.

This rise in viraemia was most prominent in the first week following immunisation, and viral titres had returned to or toward baseline by 2-3 weeks. An increase in HIV-1 titre in PBMC was seen in only one of two cases. The transiently higher levels of viraemia were not associated with a dramatic decline in the CD4 cell counts.

These findings are preliminary and should not be used to make clinical decisions about influenza vaccinations in HIV-1-infected persons. However, they should trigger more systematic studies of the acute effect of different infections or immunisations on HIV-1 replication in vivo. The impact of allogeneic exposures from blood transfusions on viral burden should also be determined.

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1. Ho DD, Moudgal T, Alam M. Quantitation of human immunodeficiency virus type 1 in the blood of infected persons. *N Engl J Med* 1989; 321: 1621-25.

Insulin autoimmune syndrome and HLA-DR4

SIR.—Dr Uchigata and colleagues (Feb 15, p 393) demonstrate that all of 27 Japanese patients with insulin autoimmune syndrome were positive for HLA-DR4 (DRB1*0406), suggesting that the development of the syndrome is linked to the presence of specific HLA class II molecules. We have evidence that this may not be so for other ethnic groups.

In 1986, a 55-year-old Swiss woman was referred to our hospital for evaluation of recurrent spontaneous hypoglycaemic attacks that usually took place 1-3 h after meal ingestion, were improved by food intake, and were precipitated by exercise. Fasting plasma total insulin (1.22 nmol/l) and proinsulin (0.48 nmol/l) were raised, and human insulin specific autoantibodies were detected both by radioimmunoassay (RIA) and by an IgG-specific ELISA. In the RIA, the patient's serum bound 49.3% of human, but only 3.7% of added pork, insulin tracer, and in the ELISA 352% (% of reference serum binding) human insulin and 2.6% pork insulin were bound. Further analysis revealed a monotypic and monoclonal human insulin autoantibody, which showed a restriction to the lambda light chain. On the basis of these findings insulin autoimmune syndrome was diagnosed.¹

HLA-typing of this patient by standard microlymphocytic toxicity tests showed the following HLA haplotype: A30, 32; B27, 62; DR 1,2. Thus, apart from the class I antigen B62, which was present in 22 of the 27 patients reported by Uchigata, our patient

had HLA antigens that were rarely seen in the Japanese group. In particular, the class II antigen DR1 was absent in the Japanese patients with insulin autoimmune syndrome, and HLA-DR2 was found in as few as 3 of them. Thus, the high prevalence of HLA-DR4 (DRB1*0406) in Japanese patients may, at least in part, indicate the high prevalence of that antigen in this ethnic group.

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1. Sklarer I, Wilkin TJ, Diaz JL, Erb P, Keller U. Spontaneous hypoglycaemia associated with autoimmunity specific to human insulin. *Diabetes Care* 1987; 10: 152-59.

Mazzotti test for onchocerciasis

SIR.—The Mazzotti test is a dangerous, provocative test that has been used to detect onchocerciasis but is condemned by the World Health Organisation expert committee on onchocerciasis.¹ It came into vogue 40 years ago as an easy way to detect this disease. In developing areas, one could always give a patient some diethylcarbamazine (DEC), and if they became sick they had "oncho". In many ways it was easier and cheaper than doing skin snips.

We now know that the use of DEC and provoking a Mazzotti reaction is not without risk. It can cause severe acute illnesses, irreversible long-term sequelae, and at times even death.²⁻⁴ These reactions are not manifestations of direct drug toxicity but are related to the death of microfilariae. However, the severity of the reaction is not closely linked to the intensity of infection, nor will every infected person have a reaction. In double-masked, controlled clinical trials, those having a Mazzotti reaction after DEC are clearly different from those who receive placebo therapy, or, for that matter, ivermectin.⁵

I was surprised to read Dr Keystone and Dr Davies' report (March 14, p 678) in which they recommend this test and describe a single-blind method for its administration. In patients for whom they consider the diagnosis of onchocerciasis, they should make a definitive diagnosis by taking a set of skin snips. If the skin snips are negative they can be repeated.⁶ The collection of skin snips is straightforward. It needs only simple equipment and is definitive when positive. The "trial by fire" approach that the Mazzotti test exemplifies seems hard to justify.

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1. World Health Organisation Expert Committee on Onchocerciasis: third report. Geneva: WHO, *Tech Rep Ser* 732, 1987: 35-61.
2. Bird AC, El-Sheikh A, Anderson J, Fagiang H. Changes in visual function and in the posterior segment of the eye during treatment of onchocerciasis with diethylcarbamazine curate. *Br J Ophthalmol* 1980; 64: 191-200.
3. Greene BM, Taylor HR, Humphrey RL. Protonuria associated with diethylcarbamazine treatment of onchocerciasis. *Lancet* 1986; i: 254-55.
4. Oomen AP. Facilities after treatment of onchocerciasis with diethylcarbamazine. *Trans R Soc Trop Med Hyg* 1969; 63: 548.
5. Greene BM, Taylor HR, Cripp EW, et al. Comparison of ivermectin and diethylcarbamazine in the treatment of onchocerciasis. *N Engl J Med* 1985; 313: 133-38.
6. Aziz MA, Diallo S, Diop IM, Lariviere M, Porta M. Efficacy and tolerance of ivermectin in human onchocerciasis. *Lancet* 1982; ii: 171-73.

*This letter has been shown to Dr Keystone and Dr Davies, whose reply follows.—ED L.

SIR.—We agree with Dr Taylor that skin snips are the method of choice to diagnose onchocerciasis and thank him for emphasising the potential dangers of the Mazzotti test, especially in patients with moderate to heavy infections. However, we do not agree that the test is without merit.

The diagnosis of early onchocerciasis is difficult. *Onchocerca volvulus* microfilariae can be absent from multiple skin snips in light or early infections.¹ Taylor and his colleagues have confirmed this by showing that the sensitivity of skin snips declines strikingly when microfilaria density drops below 1 per mg skin.² Repeat of skin snips may not always be possible because of patient intolerance of the procedure, and may be negative yet again.