LETTERS

On closer inspection

U.S. Army researchers are said to be required to follow the "Common Rule" when conducting studies on humans. Concerns are expressed by New Zealanders about exterminating rabbits with the use of a viral disease. Some in vitro alternatives (below) to the Draize rabbit eye test are said to be making progress. And the response of the immune system to tuberculin vaccination is discussed.

D. HENTGES/NORTHWESTERN UNIV.

Human Subject Research

In the ScienceScope item "Broader oversight for research on humans?" (31 Jan., p. 605) it is stated that for "Army scientists doing classified studies . . . following the Common Rule is voluntary." On the contrary, it is mandatory for the Army and all other Department of Defense researchers to follow the regulations set forth in the Common Rule "which requires that researchers obtain approval for human experiments from an institutional review board, fully inform test subjects about the risks, and obtain subjects' written consent."

As a federal agency and as part of the Department of Defense, the Army is (i) one of the 16 agencies to work for adoption of the Common Rule, and (ii) one of the agencies that adopted the Common Rule in 1991. Title 32, *Code of Federal Regulations*, Part 219, is the Common Rule for the Department of Defense. All Army and Department of Defense biomedical human subject research, whether classified or nonclassified, intramural or extramural, is subject to the Common Rule. The Army and Department of Defense are also subject to the Food and Drug Administration's oversight of investigational medical product research and use involving humans.

While it is correct that " 'there is no law saying that all research involving human subjects must have informed consent,' " the implication cannot be made that informed consent is therefore not required or obtained. In the mid-1970s, both the National Institutes of Health and the Army instituted major changes in regulations governing the oversight of human subject research. The changes, including requirements for informed consent, are enforced, and they continue to be strengthened and clarified in response to concerns of the scientific and lay community.

Dale G. Vander Hamm
Major, U.S. Army,
Chief, Human Use Review and Regulatory Affairs,
U.S. Army Medical Research and Materiel Command,
Attn: MCMR-RCQ-HR,
504 Scott Street,
Fort Detrick, MD 21702–5012, USA

■

Atopic Disease and Immunologic Response

In their report "The inverse association between tuberculin responses and atopic disorder" (3 Jan., p. 77), Taro Shirakawa *et al.* suggest that their findings are the result of a protective effect of early exposure to mycobacteria. A number of older studies of atopic dermatitis documented the same correlation, but derived an alternative conclusion—that atopic disease alters delayed hypersensitivity (*1*). Uehara (*2*) found that size of tuberculin reaction decreased and then increased in individuals as they underwent aggravation and then remission of atopic dermatitis. Although the idea that early childhood infections (and perhaps vaccination) can protect against the increasingly more common diseases of asthma and hay fever is attractive, the evidence admits of other interpretations.

David Duffy
Queensland Institute of Medical Research,
Brisbane, Qld 4029, Australia
E-mail: davidd@qimr.edu.au

References

1. H. Gudjonsson, A. Lodin, J. Modee, *Acta Dermatovena* **46**, 159 (1966); G. Rajka, *ibid.* **47**, 158 (1967); M. Forsbeck, A. Hovmark, E. Skog. *ibid.* **56**, 135 (1976).
2. M. Uehara, *Arch. Dermatol.* **113**, 1226 (1977).

In their report, Shirakawa *et al.* state (p. 77) that, in immunized schoolchildren, "[p]ositive tuberculin responses predicted

a lower incidence of asthma. . . ." These children were first immunized at 3 months of age. Although BCG elicits cellular immunity and a Th1-like response (*2*), whether it will achieve this effect may depend on the potentials of the infant's T helper cells (T_H1 and T_H2, respectively) and antigen-presenting cells (APCs).

We have found that APC maturation in healthy human infants is complex (*3*). Cord blood leukocytes (CBLs) provided APC function for generating alloantigen-stimulated interleukin-2 production. However, this activity was lost sometime between birth and 6 months and did not reappear until about 1 year (*3*). We suggested that the transient APC-presenting function seen at birth was the result of an in utero maternal cytokine influence that was lost and subsequently replaced by the infants' endogenously generated dominant T_H1-like activity (*3*). In contrast, alloantigen-stimulated interferon-γ production was impaired in CBLs (*4*). It may be relevant that infants fed cow's milk before age 3 months are at higher risk for developing T_H1-driven insulin-dependent diabetes than infants not fed cow's milk products until after the third month (*5*).

Consistent with the concept of a T_H2-like bias in infants are findings that (i) the immune potential of mice is T_H2-like during the first few days of life, before converting to a dominant T_H1-like profile (*6*); (ii) mice immunized soon after birth develop a T_H2-like response to an immunogen that is normally T_H1-dominant in adults, and they maintain T_H2-dominance even when reimmunized as adults (*7*); and (iii) human infants born to atopic mothers are more T_H2-biased than those born to atopic fathers or to nonatopic mothers (*8*). The maternal cytokine environment might be more T_H2-like in a mother with pre-existing atopy and cause this T_H2-bias.

The period between birth and approximately 3 months appears therefore to be critical for immunization, during which exposure to T_H1-inducing antigens may facilitate a reduction in subsequent T_H2-dominance. It will be important to determine whether the termination of large-scale smallpox immunization (as the result of the eradication of that disease) will further contribute to a higher incidence of asthma or other T_H2-driven conditions. Also, policy-makers should consider the possibility that some infants may be at greater risk for T_H2-driven responses and might need a different vaccine regimen from what other infants require.

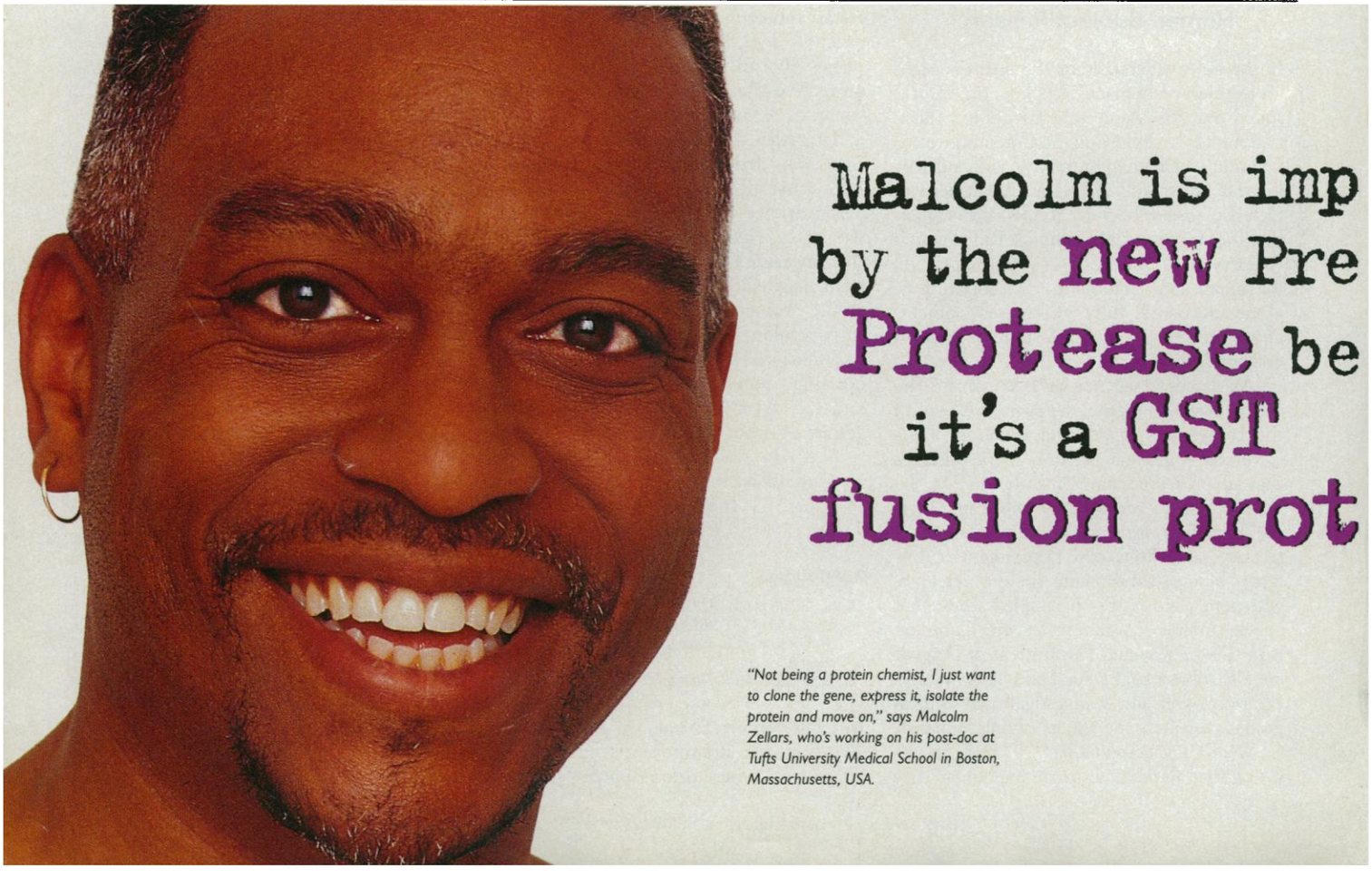
Gene M. Shearer
Claire Chougnet
Minetta S. Shearer

National Cancer Institute,
Bethesda, MD 20817–1360, USA
E-mail: shearerg@dc10a.nci.nih.gov

References

1. T. Shirakawa, T. Enomoto, S. Shimazu, J. M. Hopkin, *Science* **275**, 77 (1997).
2. W. O. C. M. Cookson and M. F. Moffatt, *ibid.*, p. 41.
3. M. Clerici, L. DePlama, E. Roilides, R. Baker, G. M. Shearer, *J. Clin. Invest.* **91**, 2829 (1993).
4. S. Taylor and Y. J. Bryson, *J. Immunol.* **134**, 1493 (1985).
5. H. C. Gerstein, *Diabetes Care* **17** (1994).
6. B. Adkins and K. Hamilton, *J. Immunol.* **149**, 3448 (1992); J. P. Ridge, E. J. Fuchs, P. Matzinger, *Science* **271**, 1723 (1996); M. Sarziotti, D. S. Robbins, P. M. Hoffman, *ibid.*, p. 1726; T. Forsthuber, H. C. Yip, P. V. Lehmann, *ibid.*, p. 1728.
7. C. Barrios *et al.*, *Eur. J. Immunol.* **26**, 1489 (1996).
8. M. P. Piccinni *et al.*, *ibid.*, p. 2293.

Response: The observed inverse association between tuberculin responses and atopic disease does admit different interpretations that will only be resolved by experimental studies. As argued in our report, we favor the conclusion that exposure to tuberculosis inhibits atopy because of the observed trends in prevalence of infectious tuberculosis, tuberculin sensitivity, and atopic disease; because of the evident mutability of tuberculin status and atopic symptoms in our population; and because of the data from animal experiments. Studies to test



"Not being a protein chemist, I just want to clone the gene, express it, isolate the protein and move on," says Malcolm Zellars, who's working on his post-doc at Tufts University Medical School in Boston, Massachusetts, USA.

whether exposure to certain mycobacteria, by inoculation, represses human atopic responses may further clarify the issue.

Shearer *et al.* raise important issues relevant to the potential and impact of immunization in childhood. Many of the tuberculin responses we observed, and their putative repression of the T_H2-mediated atopic diseases, were likely a result of natural exposure to tuberculosis rather than of immunization. Nevertheless, we share the view that study of the role and potential of childhood vaccines in deviating T_H1 and T_H2 immune profiles in an antigen-independent manner is of real interest and may be important in future attempts to prevent and restrain both atopic and autoimmune disorders in man.

J. M. Hopkin
Lung Research Laboratory,
Osler Chest Unit,
Churchill Hospital, Oxford OX 3 7LJ,
United Kingdom

■

Animal Alternatives in Germany

In his News & Comment article "Hunting for animal alternatives" (11 Oct., p. 168), Wade Roush reports on the low amount of funding of research on animal alternatives by the governments of the United States, the United Kingdom, and the Netherlands. He also reports that attempts to replace the Draize rabbit eye test have been unsuccessful to date. As head of the National German Center for the Documentation and Evaluation of Alternatives to Animal Experiments (ZEBET), I can comment on these two topics from the German point of view.

For the past 15 years, the German government has funded research to develop alternatives to the use of experimental animals at a rate of $3 to $6 million per year. ZEBET, established in 1989 as part of the German Federal Health Office, has set up an alternatives databank and operates at the national and international level with an annual budget of $400,000. Because reduction of the numbers and the suffering of experimental animals is a key political issue in Germany, the federal government has banned safety testing of cosmetic formulations since 1987.

Roush reports that the worldwide validation trial of nine in vitro alternatives to the Draize eye tests in 36 laboratories was unsuccessful (*1*). In Germany, we have since conducted a trial of two in vitro alternatives to the Draize eye test, the HET-CAM test and a cytotoxicity test, to replace the test for severely eye-irritating chemicals. The results of the trial (in 13 laboratories and on 200 chemicals) were successful and have recently been published (*2*). We have developed a sequential in vitro testing approach for classifying severely eye-irritating chemicals according to European Union regulations. We therefore have good reason to conclude that, depending on the toxicological problem to be evaluated, several in vitro assays are appropriately established to assess ocular irritancy for a given group of chemicals or for a specifically defined purpose, for example, to distinguish between severe and mildly irritating properties or between nonirritating and mildly irritating materials (*3*).

Horst Spielmann
Director, ZEBET at the BgVV,
Federal Institute for Health Protection of Consumers and Veterinary Medicine (BgVV),
Diedersdorfer Weg 1,
D-12277 Berlin, Germany

References

1. M. Balls, P. A. Botham, L. H. Bruner, H. Spielmann, *Toxicol. in Vitro* **9**, 871 (1995).
2. H. Spielmann *et al. Altern. Lab. Anim.* **24**, 741 (1996).
3. H. Spielmann, in *In vitro Methods for Pharmacotoxicological Research*, J. V. Castell and M. J. Gomez-Lechon, Eds. (Academic Press, London, 1997), pp. 265–287.