

MERTHIOLATE ALLERGY: A NATIONWIDE IATROGENIC SENSITIZATION¹

Halvor Möller

From the Department of Dermatology, University of Lund, Malmö, Sweden

Abstract. Merthiolate is an organic mercurial widely used as a preservative. Allergic eczematous reactions occur frequently in countries where merthiolate is used as a disinfectant. In Sweden where merthiolate is used mainly for incorporation into vaccines and test agents, the frequency of contact allergy to merthiolate is 3.7% in a 5-year eczema material. It is worth noting that the peak incidence occurs in the age group 20-30 years. The contact allergy may appear epicutaneously as well as intracutaneously and the histological responses agree with those of an eczematous contact allergy and of a tuberculin reaction, respectively. Cross-sensitization occurs to a few organic mercurials, but not to inorganic or metallic mercury. Merthiolate allergy is frequently correlated to the presence of the clinical picture of pompholyx, i.e. vesicular eruptions of palms and soles. Hypersensitivity to merthiolate occurs even more frequently in healthy young subjects, e.g. in 16% among military recruits. The intra-pair reaction to merthiolate among monozygotic twins is similar to that of dizygotic twins. An opportunity for sensitization to merthiolate is provided in connection with intracutaneous testing with tuberculin containing merthiolate as a preservative. It was shown experimentally that tuberculin could act as an adjuvant during sensitization to merthiolate. At present, the iatrogenic occurrence of merthiolate allergy in the Swedish population does not result in eczematous reactions—only in false-positive cutaneous tests.

Key words: Merthiolate; Mercury; Tuberculin; Pompholyx; Sensitization

Interest in patch test reactions to merthiolate was awakened in 1969 when numerous positive tests were discovered among healthy young military recruits. Initially, the nature of these reactions was poorly understood but recently it has been concluded that they are an expression of delayed hypersensitivity. The frequency and pathogenesis

of the cutaneous reactions to merthiolate have been the subject of six clinical and experimental studies (20, 21, 22, 24, 28, 33). In the following, a five-year material of merthiolate allergy is presented, with particular reference to the frequency and age distribution of this hypersensitivity as well as to the clinical picture and the occurrence of cross-sensitization.

MEDICAL USE

Merthiolate is an organic mercury compound with the following synonyms: thimerosal, thiomersal, merseptyl, mersonin, mercurothiolate, mertorgan, merfamin and others. Like many other mercurials it has shown a high antimicrobial activity (14) combined with a low acute and chronic toxicity (25, 30). Studies on the uptake and distribution of merthiolate in man and mammals have shown that it is primarily taken up in the liver and kidneys but also in the brain, skeletal muscles and other organs including skin (4, 38, 40, 42). It is mainly converted to inorganic mercury in the body (4, 40).

Merthiolate has been widely used for decades as an effective preservative in various products for medical use. Examples are vaccines for tetanus, pertussis, diphtheria, mumps and staphylococcal infections, test solutions for intracutaneous demonstration of IgE antibodies as well as of delayed allergy (tuberculin, histoplasmin, coccidioidin, mumps and candida antigens), immuno-globulin preparations, and topical medicaments (creams and lotions, eye and ear drops). The merthiolate concentration varies between 0.002 and 0.01% in these preparations. In some countries merthiolate is used as a skin disinfectant and may even be sold over-the-counter, usually as a tincture, for self-treatment of contusions, burns, etc.

¹ Presented at the IX International Congress of Allergology, October 27, 1976, Buenos Aires, Argentine.

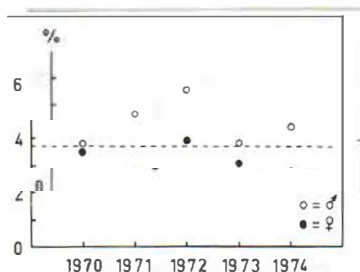


Fig. 1. The frequency (%) of patients tested) of positive patch tests to merthiolate during the years 1970–74. The mean frequency of 3.7% is indicated by a broken line.

PATCH TEST REACTIONS

Eczematous contact allergy to merthiolate has been the subject of several case reports (cf. 20). Merthiolate has also been included in battery patch test series and has thereby given varying values of positive reaction: in the USA, 13.4% in 1967 (13) and 8% in 1972 (37). From Japan a figure of 5.6% positive tests to merthiolate was reported (31), from Finland 2.0% (19) and from Denmark 1.3% (23). In East Germany, among 72 patients patch test positive to mercury only 5 were allergic to merthiolate (27).

In the Department of Dermatology in Malmö the occurrence of contact allergy to merthiolate has been registered since 1969. The results of patch testing with merthiolate during a five-year period are presented in Fig. 1. The mean frequency of merthiolate allergy has been 3.7% in a material of 600–800 patients tested per year. The figures have been slightly higher in males than in females throughout.

The 129 patients with positive patch tests to merthiolate during the years 1970–74 have been grouped according to age (Fig. 2). Merthiolate reactors may be found in all age groups but there is a predominance in young adults.

Comment. The high frequencies of merthiolate allergy reported from the USA are probably attributable to the free availability of the compound and its use as a skin disinfectant. This is not the case in Scandinavia. In Denmark, the main manufacturer of vaccines and test solutions, Statens Seruminstitut, does not even include merthiolate, but for a few exceptions. Consequently, only a few of our 129 merthiolate patients could confirm external contact with the mercurial in a corticosteroid cream, e.g. Synalar ICI-Pharma or Kenacombin

Squibb. In other words, the relevance of our test results was very low. The frequency of merthiolate allergy of 3.7% classifies merthiolate as a medium-strong allergen in our test laboratory; nickel and balsam of Peru being the leading ones, with about 9 and 7%, resp.

The present material shows an age distribution quite different from that of contact allergy in general, which usually has a peak in middle age (17). Apart from merthiolate, only nickel allergy shows a predilection for young adults (6). The predominance of the young age groups among merthiolate reactors, already pointed out in our first material (20), has recently been confirmed from Finland (19). This finding suggests that there exists in youth a probable opportunity for sensitization to merthiolate.

OTHER TYPES OF ALLERGIC REACTIONS

In the search for skin-sensitizing antibodies, delayed intracutaneous reactions to merthiolate have occasionally been observed (12, 32, 36). Even immediate reactions to merthiolate have been noted (45). One patient with an anaphylactic-type laryngeal obstruction had, however, a delayed allergy to merthiolate and had used a merthiolate-containing spray for sore throat (29).

Generalized allergic reactions to merthiolate are apparently rare. One patient with hypostatic dermatitis and contact allergy to merthiolate had a body temperature elevation after topical application of the drug (8). Another patient with similar treat-

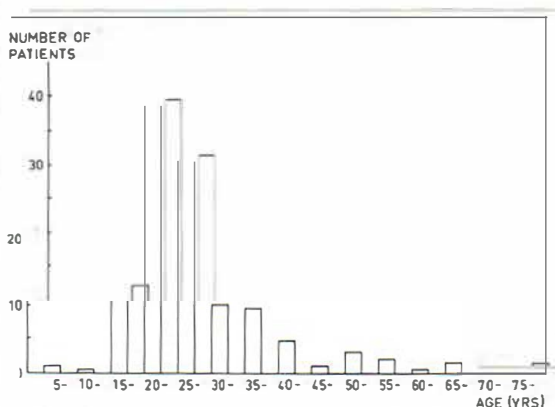


Fig. 2. The age distribution of the 129 patients with a positive patch test to merthiolate during the years 1970–74.



Fig. 3. Microscopy of a positive intracutaneous test to merthiolate. Epidermis is intact, upper corium shows a moderate edema. There is a dense inflammatory infiltrate, particularly around vessels and sweat glands, composed of small lymphocytes and some eosinophilic leukocytes (HE stain, $\times 85$).

ment experienced an enhancement of her erythroderma with temperature increase (1). Regular drug exanthems have not been reported. It is particularly noteworthy that in subjects with delayed hypersensitivity to merthiolate, no systemic reaction was observed after subcutaneous injections containing the preservative (12, 21, 36).

Comment. It might have been expected that systemic administration of merthiolate would have elicited generalized reactions in subjects with delayed allergy to merthiolate. These individuals had, however, no previous dermatitis and there was therefore little expectation of provoking an "endogenous contact eczema" (34). Furthermore, the dose of merthiolate when given as a preservative in a subcutaneous injection is very low, usually 0.05–0.1 mg.

HISTOPATHOLOGY

The clinical and histological picture of the merthiolate patch test reactions has clearly been that of an eczematous contact allergy (12, 20). The positive intracutaneous test for merthiolate has been of the Mantoux type, with erythema and infiltration (22). The microscopic picture of the intracutaneous merthiolate test has been examined in several biopsies (F. Linell). In a slightly edematous corium, particularly around vessels and sweat glands, there has been a dense inflammatory infiltrate composed of small lymphocytes with the addition of some eosinophilic leukocytes (Fig. 3). In subjects allergic to tuberculin as well as to merthiolate, the

histologic picture of the intracutaneous reactions to merthiolate, to Old Tuberculin and to Purified Protein Derivative was identical.

Comment. Histologically, both epicutaneous and intracutaneous test reactions to merthiolate fulfil the criteria for delayed hypersensitivity. Epstein (12) too noted the affinity of lymphocytes to sweat glands in the intracutaneous merthiolate reactions and he used this finding as evidence for his postulate of two different types of delayed cutaneous allergy. Our findings tally well with accepted descriptions of the tissue changes in the intracutaneous tuberculin reaction (2, 41).

CROSS-SENSITIZATION

It was reported long ago that patients with contact allergy to merthiolate reacted only to a small degree to inorganic and other organic compounds (11). These findings have been confirmed recently (16, 31), even with regard to metallic mercury (16). It was suggested instead (10) that the patients were not allergic to mercury itself but to the thiosalicylic acid part of the merthiolate molecule. In our original material of healthy recruits, 18 subjects with positive merthiolate reactions were studied for cross-sensitivity (20). Only 2 subjects reacted to thiosalicylic acid and 2 (others) to mercuric bichloride. Thus, the hypothesis of thiosalicylic acid being the allergenic determinant of the merthiolate molecule could not be confirmed.

A cross-sensitivity study has now been performed on 23 patients with positive patch tests to

Table II. *The occurrence of pompholyx in patients with merthiolate (M) allergy and in two control groups*

	Patient group		
	M	Controls, incl. nickel cases	Controls, excl. nickel cases
<i>n</i>	37	40	34
Pompholyx	15	8	3
Pompholyx %	41	20	9
Statist. difference against M group		$p < 0.05$	$p < 0.01$

the ethyl. Although immunologically different, methylmerthiolate shared the chromatographic properties of merthiolate (Fig. 4). The figure also shows that the test-positive substances were qualitatively non-related to merthiolate. This result, together with the negative tests with phenyl, etoxyethyl, and metoxyethyl mercuric compounds as well as with mercuric acetate, seem to implicate the ethylmercury radical as the allergic determinant of the merthiolate molecule. This conclusion does not, however, agree with the fairly large number of tests positive to methylmercuric dicyandiamide which cannot be explained at present.

CLINICAL PICTURE OF MERTHIOLATE DERMATITIS

It is common knowledge that contact allergy to certain topicals such as neomycin, benzocain and balsam of Peru is to a large extent correlated to the presence of hypostatic dermatitis. Otherwise such correlations between test result and clinical picture are rare. Over the years the impression has been gained, however, that hypersensitivity to merthiolate often occurs in patients with hand eczema, particularly of the pompholyx type. Therefore patients with merthiolate allergy were thoroughly examined in a controlled study with respect to their skin disease.

Patients with a positive patch test to merthiolate during the years 1969–71 were invited to a clinical examination. The total material was 47 patients but only 37 appeared for examination. Forty consecutive patients, matched as to age and sex, served as controls and were all tested during the same period of time. The controls had either a negative patch test or were positive to standard allergens other

than merthiolate. A detailed history was taken regarding the localization of skin lesions at the onset of the dermatitis and the most frequent localization during relapses. The types of skin lesions and their present localization were noted. Each individual patient was then classified, on purely clinical grounds, with one or at most two eczema diagnoses: irritant contact, allergic contact, atopic, nummular, seborrhoeic, hypostatic, pompholyx and non-specific (6). Examination and classification were carried out without knowing whether the subjects were merthiolate patients or controls.

The impression of a high frequency of pompholyx among merthiolate patients was confirmed in the present study. Of the 37 merthiolate patients examined, 15 showed this eczema type, which was seen in only 8 out of 40 controls (Table II). There were 6 patients with nickel allergy among the 40 controls; when these were excluded the frequency difference was still higher: 41 vs. 9%. Clinical examples are shown in Fig. 5.

Comment. Except for cases of eczematous contact allergy to merthiolate in patients having used merthiolate tincture as a skin disinfectant, for instance, clinical reports of merthiolate allergy have not been published. The present controlled study showed a high incidence of pompholyx in the merthiolate patients, a clinical picture that should be considered as a reaction pattern rather than a nosologic entity. Only one patient with merthiolate allergy and pompholyx type of eczema has been reported (27). It was recently shown that pompholyx is common among patients with nickel allergy (6) which fact motivated the exclusion of that category in Table II. Furthermore, pompholyx has been repeatedly connected with the oral administration of the allergen in contact dermatitis (7, 9). As mentioned above, hardly any merthiolate patients were aware of any contact with the compound. A possible relationship between test finding and clinical picture would therefore appear to be accidental ingestion of merthiolate, thereby explaining the periodic activity of the pompholyx. As far as is known, however, merthiolate does not occur in Swedish foodstuffs. Another possible eliciting agent might be a compound chemically related to and cross-sensitizing with merthiolate. Traces of methylmercury have been demonstrated over many years in Swedish fish for example (44), yet our patients did not react to methylmerthiolate. Other ethylmercuric compounds than merthiolate might of course

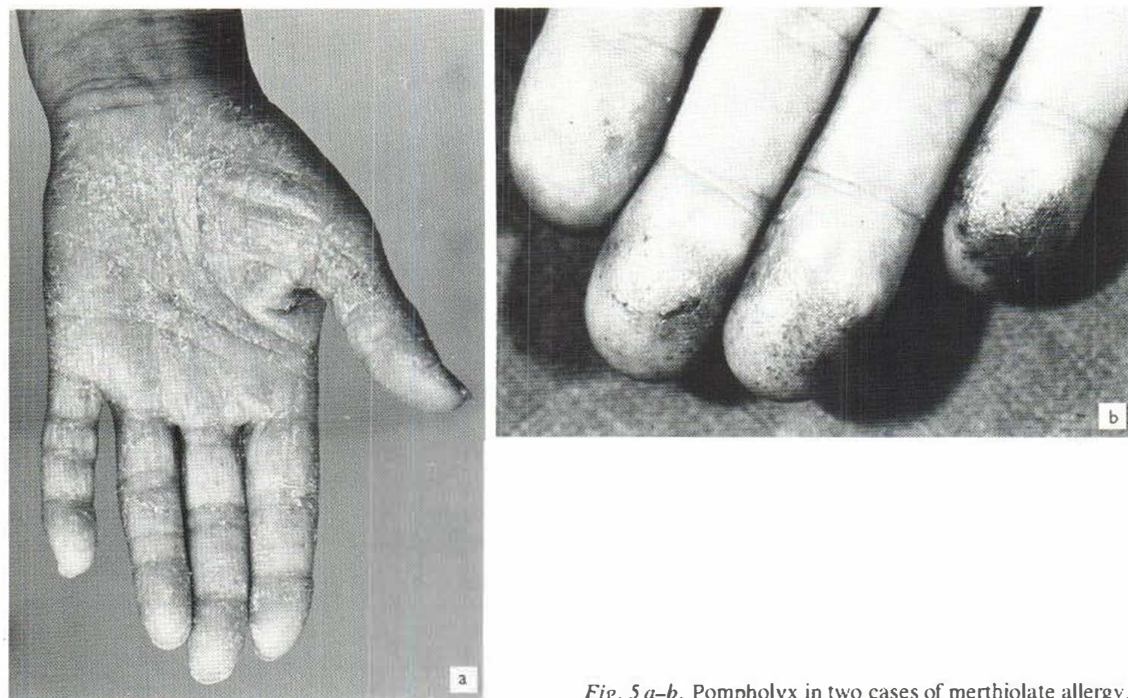


Fig. 5 a-b. Pompholyx in two cases of merthiolate allergy.

maintain the merthiolate allergy, but biotransformation from methyl to ethyl mercury does not seem to occur in man (18). Nevertheless, the association of merthiolate allergy with pompholyx is a puzzling finding that should stimulate to further search for a hidden allergen source.

HYPERSENSITIVITY TO MERTHIOLATE IN HEALTHY SUBJECTS

A remarkable finding in our first study (20) was the high frequency of 16% merthiolate reactors in a material of healthy male recruits. Positive patch tests were paralleled by positive intracutaneous

reactions (20, 21). The test-positive subjects had had no external contact with merthiolate—not even any skin disease. The result was later confirmed in another series of recruits, now using intracutaneous testing (22). Of 412 subjects tested, 15% showed a tuberculin-type reaction. This field study has now been extended to three other populations (Table III).

As may be seen from the table, the incidence of merthiolate allergy of 18% in healthy twins is very similar to that of the earlier reported recruits. A higher incidence was found in medical students (26%), and a lower one in schoolchildren (10%).

Comment. The concurring results of epicutaneous and intracutaneous testing, also observed by Epstein (12), further support the assumption of an allergic nature of the test reactions to merthiolate (15). The material with the highest incidence of merthiolate allergy, the medical students, belong to an age group corresponding to the peak frequency of merthiolate reactions in eczema patients (Fig. 2). It may thus be argued that this peak in a patient material might be a mere reflection of the prevalence in the general population. It may also explain the absence of merthiolate reactions in a material of aged patients having unselected dermatoses (20).

Table III. The result of patch testing with merthiolate in four materials of healthy subjects

Material . . .	School-children	Recruits	Twins	Medical students
Sex . . .	Both	Males	Both	Both
Median age . . .	8 y.	20 y.	20 y.	25 y.
Tested (n)	74	203	144	102
Positive (n)	7	33	26	26
Positive (%)	10	16	18	26

THE POSSIBILITY OF IDIOSYNCRASY

The high incidence of positive cutaneous tests to merthiolate in healthy subjects might be explained as being an inborn error of metabolism, rather than an allergic reaction. Thus merthiolate would, after penetration through the epidermis to the dermal connective tissue or directly deposited there, be faultily metabolized, with an ensuing inflammatory response. Such a metabolic defect might even be pharmacogenetically based.

It was found that part of merthiolate added to serum becomes rapidly bound to serum proteins (3, 33, 43). No humoral factor occurring in the serum of merthiolate reactors could be demonstrated, however (33).

The test reaction to merthiolate in pairs of monozygotic and dizygotic twins was also studied, since a pharmacogenetically determined reaction tendency would appear as a higher intrapair agreement in monozygotics. There was, however, no difference in merthiolate test reactions between monozygotic and dizygotic twins (24). As a matter of fact, this negative finding corroborated earlier conclusions that the observed test reactions were expressions of a true, delayed allergy.

OPPORTUNITY FOR SENSITIZATION TO MERTHIOLATE

The presence of delayed allergy to merthiolate in a considerable proportion of young adult Swedes must necessarily arise from some accidental sensitization when young. Lacking the possibility of epicutaneous contact with merthiolate, children and adolescents may still become sensitized during immunization and testing with agents containing merthiolate as a preservative.

Among immunological procedures that have been performed on the Swedish population during recent decades, the administration of triple vaccine (against pertussis, diphtheria and tetanus) may be considered first. The vaccine, containing merthiolate 0.01%, is given subcutaneously several times during childhood. The difficulty of inducing experimental sensitization by the subcutaneous route is notorious (26), however. In a series of 50 patients who had been given subcutaneous hyposensitization injections with merthiolate added, no one became sensitized to the preservative (36). It was later shown in a prospective study that the subcutaneous

injection of merthiolate-containing tetanus toxoid does not sensitize human subjects to merthiolate (21). Furthermore, merthiolate allergy could not be demonstrated in cases of local inflammatory reaction after subcutaneous injection of tetanus toxoid (5, 21, 45). Furthermore, poliomyelitis vaccine is given subcutaneously and routinely but does not contain merthiolate.

Merthiolate may be injected intracutaneously when skin testing with tuberculin. In Sweden, newborns are routinely vaccinated with BCG (without merthiolate). As a result thereof, and of course by any natural infection, the majority of the population gains a delayed allergy to tuberculin. Over recent decades this immunity state has been checked at the ages of 7 and 14 years in all Swedish schoolchildren, and again on taking up certain employments and educational courses, particularly in nursing and medical schools, as well as in recruits and pregnant women. Thus, in the city of Gothenburg the frequency of positive tests to tuberculin was as follows: in 7-year-olds 69%, in 14-year-olds 88%, in university students 95%, and in student nurses 97% (39). Tuberculin-negative subjects have been revaccinated and further tested. Up to 1974 the test agent in most parts of the country has been Old Tuberculin (Statens Bakteriologiska Laboratorium, Stockholm) containing merthiolate 0.01%.

EXPERIMENTAL EVIDENCE

Sensitization by the intracutaneous route in the experimental animal as well as in man has been found far more effective than by the subcutaneous or intramuscular route (26). The clinical background was as follows: 1) an intracutaneous injection of the possible sensitizer, merthiolate, repeated a couple of times at an early age; 2) the sensitizer given together with tuberculin, most likely resulting in a positive tuberculin test, i.e. an inflammatory reaction containing immunocompetent lymphocytes. Thus, the conditions for developing a sensitization to merthiolate were highly suitable. Could even tuberculin, a bacterial culture filtrate, work as an adjuvant like Freund's agent, a bacterial emulsion?

Sensitization to merthiolate was easily induced by the "guinea pig maximization test" (28). In tuberculin-positive guinea pigs, more animals became sensitized to merthiolate when tuberculin was added to merthiolate during the sensitization proce-

ture than when merthiolate was given alone. Thus, the hypothesis was confirmed that tuberculin can act as an adjuvant. Thus, a reasonable explanation was provided for the high incidence of positive cutaneous tests to merthiolate in the younger Swedish population.

PRACTICAL IMPLICATION

The various findings described above all lead to the conclusion that the positive patch test reactions to merthiolate in the Swedish population imply an eczematous contact allergy to this compound, though usually not to mercury as such. In subjects without external contact with merthiolate, such allergy appears to be of minor clinical importance. Should, however, merthiolate become introduced in this country as, say, a household disinfectant, then an accumulation of eczematous reactions would probably follow.

At present, and probably for several years ahead, the high incidence of delayed allergy to merthiolate in the Swedish population will give rise to false-positive test reactions when applying allergens containing merthiolate as a preservative (22).

REFERENCES

1. Amblard, P., Dugois, P., de Bignicourt, B. & Imbert, R.: Poussées thermiques intenses chez une érythrodermique déclenchées et entretenues par des applications d'antiseptiques liquides mercuriels incolores. *Lyon Méd* 220: 205, 1968.
2. Bandmann, H.-J. & Szarmach, H.: Beitrag zur Histopathologie der intracutan ausgelösten Tuberkulinreaktion. I. Problemstellung und deren Begründung an Hand der bisher veröffentlichten Untersuchungen. *Arch Klin Exp Derm* 218: 1, 1964.
3. Batty, I., Harris, E. & Gasson, A.: Preservatives and biological reagents. In *International Symposium on Preservatives in Biological Products*, San Francisco, 1973. Develop Biol Standard, vol. 24, p. 131. S. Karger, Basel 1974.
4. Blair, A. M. J. N., Clark, B., Clarke, A. J. & Wood, P.: Tissue concentrations of mercury after chronic dosing of squirrel monkeys with thiomersal. *Toxicology* 3: 171, 1975.
5. Christensen, R. E.: Discussion to Benjamini, E.: Potential antigenic and haptenic properties of preservatives. In *International Symposium on Preservatives in Biological Products*, San Francisco, 1973. Develop Biol Standard, vol. 24, p. 194. S. Karger, Basel, 1974.
6. Christensen, O. B. & Möller, H.: Nickel allergy and hand eczema. *Contact Dermatitis* 1: 129, 1975.
7. Christensen, O. B. & Möller, H.: External and internal exposure to the antigen in the hand eczema of nickel allergy. *Contact Dermatitis* 1: 136, 1975.
8. Cogswell, H. D. & Shoun, A.: Reaction following the use of Tr. merthiolate. *Ariz Med* 5: 42, 1948.
9. Ekelund, A.-G. & Möller, H.: Oral provocation in eczematous contact allergy to neomycin and hydroxy-quinolines. *Acta Dermatovener (Stockholm)* 49: 422, 1969.
10. Ellis, F. A.: The sensitizing factor in merthiolate. *J Allergy* 18: 212, 1947.
11. Ellis, F. A. & Robinson, H. M.: Cutaneous sensitivity to merthiolate and other mercurial compounds. *Arch Dermatol Syph* 46: 425, 1942.
12. Epstein, S.: Sensitivity to merthiolate: a cause of false delayed intradermal reactions. Clinical and histological investigations. *J Allergy* 34: 225, 1963.
13. Epstein, E., Rees, W. J. & Maibach, H. I.: Recent experience with routine patch test screening. *Arch Dermatol* 98: 18, 1968.
14. Farber, J. F. & Rench, M.: Testing of preservatives in biological products. In *International Symposium on Preservatives in Biological Products*, San Francisco, 1973. Develop Biol Standard, vol. 24, p. 213. S. Karger, Basel, 1974.
15. Fregert, S. & Rorsman, H.: Allergy to chromium, nickel and cobalt. *Acta Dermatovener (Stockholm)* 46: 144, 1966.
16. Fregert, S. & Hjorth, N.: Increasing incidence of mercury sensitivity. The possible role of organic mercury compounds. *Contact Dermatitis Newsletter* 5: 88, 1969.
17. Fregert, S., Hjorth, N., Magnusson, B., Bandmann, H.-J., Calnan, C. D., Cronin, E., Malten, K., Meneghini, C. L., Pirilä, V. & Wilkinson, D. S.: Epidemiology of contact dermatitis. *Trans St Johns Hosp Dermatol Soc* 55: 17, 1969.
18. Goldwater, L. J.: Mercury in the environment. *Sci Am* 224: 15, 1971.
19. Hannuksela, M., Kousa, M. & Pirilä, V.: Allergy to ingredients of vehicles. *Contact Dermatitis* 2: 105, 1976.
20. Hansson, H. & Möller, H.: Patch test reactions to merthiolate in healthy young subjects. *Br J Dermatol* 83: 349, 1970.
21. — Cutaneous reactions to merthiolate and their relationship to vaccination with tetanus toxoid. *Acta Allergol (Kbh)* 26: 150, 1971.
22. — Intracutaneous test reactions to tuberculin containing merthiolate as a preservative. *Scand J Infect Dis* 3: 169, 1971.
23. Hjorth, N.: Sensitivity to organic mercury compounds. *Contact Dermatitis Newsletter* 1: 15, 1967.
24. Holst, R. & Möller, H.: Merthiolate testing in twins. *Contact Dermatitis* 1: 370, 1975.
25. Kirschstein, R. L.: Toxicology and carcinogenicity of preservatives used in the preparation of biological products. In *International Symposium on Preservatives in Biological Products*, San Francisco, 1973. Develop Biol Standard, vol. 24, p. 203. S. Karger, Basel, 1974.
26. Kligman, A. M.: The identification of contact al-

- lergens by human assay. II. Factors influencing the induction and measurement of allergic contact dermatitis. *J Invest Dermatol* 47: 375, 1966.
27. Loechel, I. & Zschunke, E.: Zur Aktualität der Quecksilberallergie. *Dermatol Monatsschr* 157: 570, 1971.
 28. Magnusson, B. & Möller, H.: The adjuvant effect of tuberculin in experimental sensitization to merthiolate. *Arch Dermatol Forsch*, unpubl. data.
 29. Maibach, H.: Acute laryngeal obstruction presumed secondary to thiomersal (merthiolate) delayed hypersensitivity. *Contact Dermatitis* 1: 221, 1975.
 30. Mason, M. M., Cate, C. C. & Baker, J.: Toxicology and carcinogenesis of various chemicals used in the preparation of vaccines. *Clin Toxicol* 4: 185, 1971.
 31. Masuda, T., Honda, S., Nakauchi, Y., Ito, H., Kinoshita, M., Harada, S., Yaoita, H. & Mizoguchi, M.: Patch test. The data at the Allergy Clinic of Department of Dermatology of Tokyo University Hospital for the past three years. *Jpn J Dermatol, Ser. B*, 80: 133, 1970.
 32. Mizutani, H.: Hypersensitivity to thimerosal. *N Engl J Med* 289: 1424, 1973.
 33. Möller, H.: Chemical assay of merthiolate after incubation with serum from subjects with positive patch tests to merthiolate. *Dermatologica* 145: 280, 1972.
 34. Pirilä, V.: Endogenic contact eczema. *Allerg Asthma-forsch* 16: 15, 1970.
 35. — Chambertest versus lapptest. *Förhandlingar vid Nordisk Dermatologisk Förenings 20:e möte* (ed. N. Thyresson & Ö. Hägermark), p. 43. Almqvist & Wiksell, Uppsala 1974.
 36. Reisman, R. E.: Delayed hypersensitivity to merthiolate preservative. *J Allergy* 43: 245, 1969.
 37. Rudner, E. J., Clendenning, W. E., Epstein, E. et al.: Epidemiology of contact dermatitis in North America: 1972. *Arch Dermatol* 108: 537, 1973.
 38. Saito, M., Osono, T., Watanabe, J., Yamamoto, T., Takeuchi, M., Ohyagi, Y. & Katsunuma, H.: Studies on Minamata disease. I. Establishment of the criterion for etiological research in mice. *Jpn J Exp Med* 31: 277, 1961.
 39. Sievers, O.: Sammanfattande rapport beträffande Dispensärcentralens i Göteborg undersökningar med PPD tuberkulin under läsåret 1967–1968. Stencil 1969.
 40. Suzuki, T., Takemoto, T.-I., Kashiwazaki, H. & Miyama, T.: Metabolic fate of ethylmercury salts in man and animal. 4th Int Conf Environ Toxic. Mercury, Mercurials and Mercaptans. Proc Publ Rochester, p. 209, 1973.
 41. Turk, J. L.: The histology of delayed hypersensitivity reactions in the skin. In *Delayed Hypersensitivity* (ed. A. Neuberger & E. L. Tatum), p. 46. North-Holland Publishing Company, Amsterdam, 1967.
 42. Urakubo, G. & Kido, Y.: Rectal absorption and distribution of ^{203}Hg -labeled thimerosal. *Chem Abstr* 65: 11140, 1966.
 43. Watanabe, K. & Izumi, T.: Separation of water-nondialyzable thimerosal in serum by cysteine equilibrium dialysis. *Jpn J Med Sci Biol* 26: 179, 1973.
 44. Westöö, G.: Determination of methylmercury compounds in foodstuffs. II. Determination of methylmercury in fish, egg, meat, and liver. *Acta Chem Scand* 21: 1790, 1967.
 45. White, W. G., Barnes, G. M., Barker, E., Gall, D., Knight, P., Griffith, A. H., Morris-Owen, R. M. & Smith, J. W. G.: Reactions to tetanus toxoid. *J Hyg (Camb)* 71: 283, 1973.

Received September 30, 1976

H. Möller, M.D.
Department of Dermatology
General Hospital
S-214 01 Malmö
Sweden