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CELLULAR CHANGES IN THE PSORIATIC EPIDERMIS

IX. *Neutron Activation Analysis of Mercury in Patients Topically Treated with Ammonium Mercuric Chloride*

A. Frithz

From the Department of Dermatology, Karolinska Sjukhuset, and the Department of Forest Genetics, Royal College of Forestry, Stockholm, Sweden

Abstract. Samples of skin, blood and urine from psoriatic and normal males treated for 7 days with an ointment containing 5% ammonium mercuric chloride have been subjected to neutron activation analyses in attempt to study the transepidermal absorption of mercury. A highly increased mercury content was found in skin with a considerable difference between normal and psoriatic epidermis and corresponding elevated values in blood with the same difference. The conjectured causal mechanism is discussed. Estimation of methyl mercury in skin is reported.

Psoriasis is a chronic and fluctuant disease, the clinical features of which were very well described by Willan (26) in 1808. The course of the disease is wellknown and the microscopic and submicroscopic structures have been studied by several authors (6, 10, 11, 13, 15, 25). The treatment of psoriasis is almost entirely confined to local applications. Applications of mercury compounds to the psoriatic skin have been used since before the days of Willan. Ammoniated mercury, which seems to have the least percutaneous passage, has been extensively employed in treatment. A multiplicity of studies on the problems concerning the presence of mercury in nature and living organisms has been performed. In this work, literature of interest regarding the present investigation is referred to. The toxic effects of mercury and mercury compounds, due to transepidermal absorption, to ingestion and to inhalation, have been investigated from the viewpoint of environmental health (1, 2, 3, 4, 5, 8, 16, 22, 23).

Studying the distribution of mercury within the body tissues, following parenteral administration of mercury compounds, Berlin & Ullberg (2, 3, 4) found a widespread distribution in different organs with greatest concentrations in the kidneys. Friberg (8) reported that methyl mercury was more firmly bound to the tissues than was inorganic mercury and possessed a greater power of concentration to the blood–brain barrier. Bäckström (7), using autoradiography, scintillation counting and zone electrophoresis, studied the localization of mercury in different tissues of Japanese quail after parenteral or peroral administration of certain mercury compounds. Methyl mercury was characterized by an even distribution of mercury in most organs, while phenyl mercury, methoxyethyl mercury and inorganic mercury, showing mutual similarities, had the kidneys, the liver and the yolk as main target tissues. Keratinized structures strongly concentrated mercury irrespective of the mercurial given.

Investigating the distribution and excretion after injection of mercuric nitrate, phenyl mercuric acetate and methyl mercuric hydroxide, Swensson et al. (22) found that the two organic compounds are, to a large extent, bound to the erythrocytes, while the inorganic compound was carried in the plasma. Sollman (21) considers that mercurous compounds are oxidized and that the mercuric salts form soluble compounds with proteins, sodium chloride, blood and tissue fluid alkalis. Absorbed mercury leaves the blood and is excreted in the urine and faeces in amounts dependent on the compound used.

Some metabolic properties of methyl mercury and dimethyl mercury were studied in mice by Östlund (28) using radioactive substances. Whole-body radioactive measurement technique showed that the total excretion of mercury per time unit

after a single injection of methyl mercury is approximately proportional to the simultaneous mercury concentration in the body at any given time. The excretion rate was slower when larger doses were given which may be due to an increasing mercurial inhibition of sulphydral enzymes participating in the excretion processes or due to the binding of methyl mercury in the tissues to sites with different chemical properties.

Presumably the excretion of mercury occurs by tubular rather than by glomerular filtration because of the binding of mercury to the -SH groups of plasma proteins, particularly albumin (1, 5). Lundgren et al. (16), investigating the distribution of mercury in human blood and plasma and the excretion in urine after exposure to different mercury compounds, found that differences of the distribution of mercury in blood, and the rate of excretion depend on the kind of mercurials used. Applying neutron activation analysis in a group of non-exposed persons, the content of mercury was determined to 6–12 ng/g in whole blood and 3–6 ng/g in plasma. Tejning found an average of 10.1 ng/g in blood corpuscles and 2.3 ng/g in plasma in a material of 83 non-exposed persons (23). Poisonous absorption of mercury in patients treated with ointments containing mercury compounds has been reported, and Young (27) found symptoms or signs of mercury poisoning in about 50% in a group of 70 psoriatic patients. Rothman (17) has stated that the poor absorption of mercury from ammoniated mercury preparations through intact skin in contrast to the rapid absorption from, for example, the vagina is not a question of the presence of a barrier in the skin but possibly of the pH of the environment.

To explore the distribution of absorbed mercury after treatment with the ammoniated mercury ointment used in the management of psoriasis, Hg^{203} determination was performed after neutron irradiation of samples of skin, blood and urine from treated psoriatic patients.

The present study of neutron activation analyses of mercury in skin and body elements was also undertaken to investigate whether differences of mercury content in treated and non-treated materials might correspond to the reported enhancement in electron density of certain submicroscopic epidermal cellular structures (9, 14) treated in vivo with the ointment containing ammonium mercuric chloride.

MATERIAL AND METHODS

Eight male adult hospitalized patients, aged 32–64 years, with clinically manifested and histologically verified psoriasis with ubiquitous lesions were selected for the present study. The skin area involved by psoriatic lesions was estimated to about 50–60%. Hypersensitivity to mercury was excluded by patch testing. Improvement of all patients was obtained. None of the patients had previously received any treatment with substances containing mercury compounds or had had any contact with mercurials in their work. No ingestive pathways of mercury or other exposure to mercury, for instance dental treatment, were detectable.

After removal of scales with 2% salicylic acid in vaseline, the psoriatic lesions of the patients were treated twice daily for 7 days with an ointment containing 5% ammoniated mercury in a base consisting of 20% adeps lanae and 80% vaseline. Specimens of skin for determination of mercury were obtained by punch biopsies. The blood was separated into erythrocytes and plasma, and urine samples were collected. Specimens were taken in the morning the day after treatment had finished. All instruments and containers were sterilized and prepared to avoid contamination by mercury. After preparation, the material was transferred to small tubes of quartz for the activation analysis. After neutron irradiation of the samples in a nuclear reactor[1] followed by a chemical separation based mainly on distillation, mercury was determined by γ-spectrometry with a sensitivity limit of about 5×10^{-4} ppm (20).

The content of methyl mercury compound was determined by gas chromatography according to Westöö (24). The specimens were prepared in hydrobromic acid, benzene and cystein solutions for the gas chromatography as described by Kitamura et al. (12).

RESULTS

In all specimens from patients treated with ammoniated mercury ointment elevated values of mercury were found. There was a considerable variation in the amount of mercury in skin biopsies, ranging from 56 500 ng/g to 880 000 ng/g. Analyses of blood samples revealed similar ratios. No definite correlation to mercury content of the skin or urine could be established. The relations between contents of mercury in plasma, erythrocytes and whole blood is inconstant. The amount of mercury in urine ranges from 59 ng/g to 330 ng/g and roughly varies with the plasma content (Table I).

Samples from two healthy volunteers revealed highly elevated values of mercury in treated skin and moderately elevated in erythrocytes but only a slightly increased amount of mercury in plasma, whole blood and urine. The difference in mercury

[1] Performed at Isotoptekniska Laboratoriet, Stockholm, Sweden.

Table I. *Neutron activation analyses*

Case	Skin (ng/g)	Blood (ng/g)	Erythrocytes (ng/g)	Plasma (ng/g)	Urine (ng/g)
Psoriasis treated with ammonium mercuric chloride					
1	56 500	355	200	375	190
2	115 000	85	94	100	142
3	330 000	234	295	236	59
4	692 000	260	256	330	66
5	880 000	450	435	515	330
Normal individuals treated with ammonium mercuric chloride					
1	18 000	19	18	12	11
2	26 000	20	15	15	12
Untreated psoriatic skin					
1	187				
2	102				
Untreated normal skin					
1	47				
2	61				

content between normal and psoriatic skin is marked after treatment, while similar amounts of mercury are found in untreated psoriatic and normal skin (Table I). According to gas chromatographic analyses of the ammoniated mercury ointment used, the part of methyl mercury was 8 ng/g. In psoriatic lesions treated with the ointment 55–70 ng/g methyl mercury was found, in blood 2–5 ng/g and in urine less than 1 ng/g (Table II).

DISCUSSION

A considerable increase in the mercury content was noted in the various investigated specimens. The amount of mercury applied to the skin is difficult to calculate but can be estimated as about 2–4 g/m^2 mercury of the psoriatic lesions. As is known from autoradiographic analysis, mercury passes diffusely through the epidermis and systemic absorption begins after 8 hours (18), thus the punch biopsies for skin samples were performed after about this interval. Scott (18) has pointed out that a much higher proportion of the applied mercury is retained in the psoriatic than in normal skin. According to this author, the retention of mercury in the psoriatic epidermis is about 50%. Mercury was found (18) evenly spread throughout all the epidermal layers. The high content of mercury in psoriatic skin, estimated by neutron activation analyses, is therefore anticipated.

The remarkable dissimilarity between the quantities of mercury in treated and non-treated normal and psoriatic skin amply supports the electron microscopic results (9, 14) of increase in density after treatment in vivo with the ointment containing ammonium mercuric chloride, reflecting the precise intracellular distribution of mercury.

The opinion expressed by I. Silberberg (19) that "physiochemical changes induced by the mercury at those sites", or that "the mercury itself could have induced this change directly passing through these areas or by affecting these compounds indirectly" seems hard to perceive in view of the obtained heavy metal-like electron absorption (9, 14) and the results obtained by neutron activation analyses.

In the patients all the psoriatic lesions were treated with ammoniated mercury ointment. Because of the widespread distribution it was impossible to estimate the age of the individual lesions with any degree of certainty, thus lesions in all developmental stages are represented. As is known from submicroscopic studies on psoriatic lesions of different age, the development of the aberrant cellular differentiation is a dynamic process with great disparities related to age (13). Changes in the differentiation, and consequently in available reacting groups, might be a factor connected with the retention of mercury. The development of parakeratosis is individual and is able to influence the absorption of mercury. In normal skin a relatively low percentage of mercury is retained in the epidermis, about 10% according to autoradiographic analysis (18).

In spite of the absence of evidence permitting certain estimation of the intracellular chemical binding of mercury, it is conceivable that—without excluding other possibilities—the supposed

Table II. *Methyl mercury analyses in skin*

Case	Treated psoriasis (ng/g)	Untreated psoriasis (ng/g)	Normal skin (ng/g)
1	70	5	4
2	55	2	2

occurrenec of non-reactive -SH groups in normal skin (18) could be related to the observed differences between normal and psoriatic skin. An impairment of the barrier function in the psoriatic lesions, attributed to normal epidermis, is a factor that can play an important rôle in eliciting this difference.

Electron microscopic studies on the intracellular distribution of mercury (9, 14), indicated an affinity of mercury to certain submicroscopic structures. It also seems justifiable to assume that the higher number of certain of these structures (cf. 9, 14) and the larger size of the psoriatic epidermal cells might provide increased possibilities of intracellular mercury association as compared with those of normal epidermal cells. The inconstant relationship between the concentration of mercury in skin and blood is disconcerting.

A dependency on the differences in age and parakeratosis formation cannot, however, be denied.

The discrepant blood values of mercury of normal and psoriatic individuals when correlated to the amounts in skin might be a reflection of vascularity differences between normal and psoriatic skin. So far, however, the influence of other functional mechanisms cannot be excluded.

Mercury compounds have been studied with regard to their carriage in blood. Concerning ammonium mercuric chloride, no corroborative differences in concentrations between plasma and erythrocytes can be established from this material. However, it is interesting to note a tendency to higher concentration in plasma than in erythrocytes. The distribution varies, depending on which mercury compound the individual is exposed to. Until recent years, the concentration of mercury in urine has been used in attempt to control the exposure and uptake of mercury because of methodological difficulties in determining small amounts of mercury in blood. Consequently no comparable estimations are available for the mercury compound used.

Estimation of the proportion of methyl mercury in psoriatic skin treated with ammonium mercuric chloride revealed values which closely correspond to the amount of methyl mercury applied to the skin. No active formation of methyl mercury seems to take place. A corresponding difference concerning the normal skin was noted.

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Anders Frithz, M.D.
Department of Dermatology
Karolinska sjukhuset
S-104 01 Stockholm 60
Sweden