

Repeated Open Application Tests with Methyl dibromoglutaronitrile in Dermatitis Patients with and without Hypersensitivity to Methyl dibromoglutaronitrile

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Background: Contact allergy to and allergic contact dermatitis from methyl dibromoglutaronitrile (MDBGN) have been frequently reported since the 1990s.

Objective: This study was initiated to help determine the optimal test preparation for MDBGN and to help determine the clinical relevance of such a contact allergy.

Method: In 38 patients with positive (32 patients) or doubtful (6 patients) patch-test reactions to at least one of four test preparations of MDBGN in petrolatum at 1.0%, 0.5%, 0.3%, and 0.1% (all weight per weight [w/w]), a repeated open application test (ROAT) with moisturizers with and without MDBGN at 0.03% w/w was conducted on patients' upper arms for a maximum of 4 weeks. Seven patients not hypersensitive to MDBGN served as controls and went through the same procedure.

Results: Nineteen (59.4%) of the 32 MDBGN-allergic patients developed a positive ROAT result on the arm on which the moisturizer containing MDBGN was applied whereas no patients with doubtful or negative reactions to MDBGN reacted ($p = .002$). A statistically significant association was found between the patch-test reactivity and the outcome of the ROAT ($p < .001$).

Conclusion: Patch testing with MDBGN at 0.3% and 0.1% will miss clinically relevant patch-test reactions to MDBGN.

CONTACT ALLERGY TO THE PRESERVATIVE methyl dibromoglutaronitrile (1,2-dibromo-2,4-dicyanobutane) has been frequently reported since the 1990s.¹⁻⁸ In addition, allergic contact dermatitis from methyl dibromoglutaronitrile (MDBGN) in products such as toiletries used in the domestic setting and in the workplace, moist toilet paper, ultrasonic gels, and detergents and glues has also often been reported.^{1,9-24} Thus, MDBGN has been a strong candidate to be introduced in the European Standard patch-test series, and a recommendation to include it has recently been published.²⁵ However, there has been no agreement on which test preparation, with regard to vehicle and concentration, should be used to define contact allergy to MDBGN. As the purpose of patch testing with a standard test series is to find individuals with a clinically relevant contact allergy,²⁶ two studies have been performed within the European Environmental and Contact

Dermatitis Research Group to help determine the optimal patch-test preparation for MDBGN.^{27,28} With this background, the inclusion of MDBGN in the European Standard patch-test series was more than expected. A previous repeated open application test (ROAT) with and without moisturizers containing MDBGN showed that relevant allergic reactions could be missed if patch testing was done with MDBGN concentrations of less than 0.5%.²⁸ This study is a revised ROAT performed as a controlled, randomized, and double-blind study, in contrast to the previous study, which did not include patients who were not hypersensitive to MDBGN.

Materials and Methods

Patients

The ROAT was conducted on 32 dermatitis patients who were hypersensitive to MDBGN when patch-tested with four concentrations of MDBGN, (1.0% w/w, 0.5%, 0.3%, and 0.1%). Seven dermatitis patients who were not hypersensitive to MDBGN when patch-tested with the same concentrations constituted the controls and also

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participated in the ROAT. Another 6 dermatitis patients who did not have contact allergic reactions to MDBGN but had one or more doubtful reactions to one or more of the four concentrations of MDBGN also went through the ROAT. None of the 45 patients had reacted to paraben mix in the standard series. Informed written consent was obtained from each participant, and the ethical committee at Lund University approved the study.

Chemicals

MDBGN from the same batch (obtained from Schülke and Mayr, Hamburg, Germany) was used for both the patch testing with the four MDBGN preparations and the ROAT. The pharmacy at the University Hospital in Malmö prepared two moisturizers that were identical (petrolatum, sorbitan oleate, and water) but for the preservative. One moisturizer was preserved with MDBGN at 0.03% w/w, and the other was preserved with methylparaben 0.1% w/w and propylparaben 0.2% w/w.

Patch Testing

All 45 participants were patch-tested with four concentrations of MDBGN in petrolatum at 1.0%, 0.5%, 0.3% and 0.1% (all w/w) at the start of the study. The Finn Chamber technique (Epitest Ltd Oy, Tuusula, Finland) was used, and tests were placed on the upper back for 48 hours, secured with Scanpor tape (Norgesplaster A/S, Vennesla, Norway). The tests were read on day 3 according to International Contact Dermatitis Research Group (ICDRG) criteria²⁹ by one of the investigators, who did not know whether the patients were test patients or controls and who did not see the participants for the ROAT readings. Right after the patch testing, the ROAT was started.

Repeated Open Application Test

The patients were instructed to use the moisturizers twice daily on the ventral aspects of the upper arms, where a square measuring 5×5 cm was marked. Each patient received a pair of moisturizers with and without MDBGN. The pairs were numbered, and the allocation of the moisturizer to be applied to the respective arm was done in a randomized way. The moisturizer to be applied to the right upper arm was marked with a blue tape whereas a red tape was used for the moisturizer to be applied to the left arm. The patients were instructed to use an approximately 0.5 cm-long string of the cream for each application

(35 mg on $25 \text{ cm}^2 = 1.4 \text{ mg/cm}^2$). The study period was 4 weeks unless terminated earlier because of a positive ROAT result or at the patient's request. Inspection of the upper arms was done before the ROAT on the first day and then after 1, 2, 3, and 4 weeks or at the request of the patient. An eczematous reaction was sought and was considered to be positive if there was at least an erythematous infiltration with or without papules and/or vesicles covering at least 25% of the marked area.³⁰ The patients were encouraged to continue the applications if there was only an erythematous reaction without infiltration or an eczematous reaction covering less than 25% of the area. The code was broken after termination of the study.

Statistical Calculations

The Fisher exact test was used to compare the ROAT outcomes for MDBGN-allergic patients and for controls. The McNemar test was used to compare the number of patient arms with a positive ROAT result for the moisturizers with and without MDBGN. This comparison was also done in those who had a negative or doubtful patch-test reaction to the 0.1% and 0.3% concentrations but a positive reaction to 0.5% and/or 1.0% concentrations. The Cochran-Armitage trend test was used to investigate any association between the individual patch-test reactivity (PTRL), defined as the lowest patch-test concentration of MDBGN to yield a positive reaction (ie, at least a + reaction according to ICDRG guidelines²⁹), and ROAT outcome, defined as the number of days until a positive ROAT result appeared. A *p* value less than or equal to .05 was considered statistically significant.

Results

Of the 32 MDBGN-hypersensitive patients, 19 (59.4%) developed a positive response to the ROAT. None of the 6 patients with one or more doubtful reactions to MDBGN or any of the 7 patients with negative reactions to MDBGN had a positive ROAT response ($p = .002$, Fisher exact test, two sided). For all 19 ROAT-positive patients except one (who tested positively with both moisturizers), there was only a positive ROAT response for the moisturizer with MDBGN ($p < .001$, McNemar test, two sided). Within the first week, 9 patients responded positively. Five more patients responded positively in the second week, 3 more in the third week, and 2 more in the fourth week. When the same comparison was made for those who had a negative or doubtful patch-test reaction to 0.1% and 0.3%

concentrations but a positive reaction to 0.5% and/or 1.0% concentrations, 4 patients had a positive ROAT response to the moisturizer with MDBGN but not to the moisturizer without MDBGN, and 1 patient had a positive ROAT response to both moisturizers ($p = .06$; McNemar test, one sided).

Six participants had doubtful patch-test reactions to MDBGN at 0.3% and positive reactions to both of the higher concentrations; in 50% of patients, a positive ROAT reaction developed on the arm to which the moisturizer with MDBGN had been applied. Figure 1 shows the relationship between the outcome of the ROAT with regard to the number of days until a positive ROAT response appeared and the respective PTRL, including patients with doubtful reactions ($p < .001$, Cochran-Armitage trend test), and Figure 2 shows the distribution of positive and negative ROAT results for the respective PTRLs, including doubtful reactions. Two patients only reacted with an allergic reaction to 1.0% (the highest test concentration) but still had a positive ROAT reaction to the moisturizer with MDBGN. One of these two patients also had a positive ROAT reaction to the moisturizer without MDBGN.

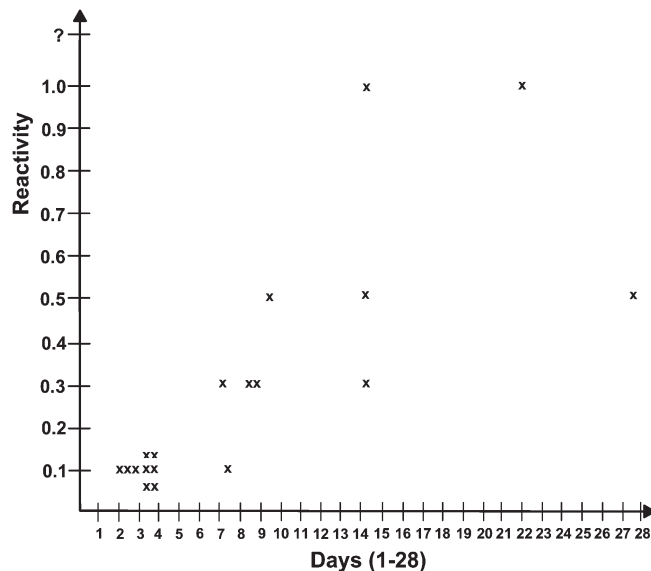


Figure 1. Relationship between the outcome of the repeated open application test (ROAT) with regard to the number of days until a positive ROAT response appeared and the respective patch-test reactivity level (0.1–1.0%) in 32 patients with allergic reactions to methyl dibromoglutaronitrile and 6 patients with doubtful (?) reactions. Nineteen patients had a positive ROAT result (seen here), and 19 had a negative ROAT result.

Discussion

ROATs and other types of use tests can be used for both (1) individual cases, to help assess the clinical relevance of a positive or doubtful reaction to a sensitizer present in a product used by the patient but that yielded a negative response on the product patch test and (2) groups of hypersensitive patients undergoing ROATs with a product containing the sensitizer. For the individual ROAT, a positive test result does not say anything about the nature of the reaction (ie, the eczematous response can be either allergic or irritant). On the other hand, when a ROAT is scientifically designed and conducted, information can be gained about both the general clinical relevance and the nature of the ROAT reactions. The present study gives information on both and can be used as an instrument to help determine the optimal patch-test preparation of MDBGN. The study demonstrates the clinical relevance of contact allergy to MDBGN traced by patch-testing with petrolatum preparations of MDBGN at 0.1 to 0.5%. A dose-response relationship for patch-test reactivity and ROAT outcome (see Fig 1) was also demonstrated, which gives further support to the significance of the demonstrated positive patch-test reactions.

When ROATs are conducted in groups of individuals with contact allergy, false-positive reactions and other irritant reactions can be recognized by the use of appropriate controls, which were used in this study. The statistically significant difference between the outcomes of the ROATs for patients and controls without allergic reactions to MDBGN (but sometimes with doubtful reactions) further supports this. This study demonstrates that a person who reacts positively to MDBGN at concentrations equal to or lower than 0.5% should avoid using moisturizers preserved with MDBGN, to avoid contact dermatitis relapses. This is supported by the fact that the concentration of 0.03% of MDBGN used in this study (chosen because chemical analysis of several hundred leave-on products on the Swedish market by the Department of Occupational and Environmental Dermatology in Malmö had shown 0.03% to be the mean concentration of MDBGN in products preserved with this preservative [personal communication: Birgitta Gruvberger March 15, 2005]) is less than the highest concentration allowed in leave-on products (0.1%, except for sunscreen products with 0.025%). We have for many years advised our MDBGN-hypersensitive patients to also avoid rinse-off products preserved with MDBGN. This advice is supported by the result of a recent study in which a liquid soap preserved with MDBGN caused eczema in allergic

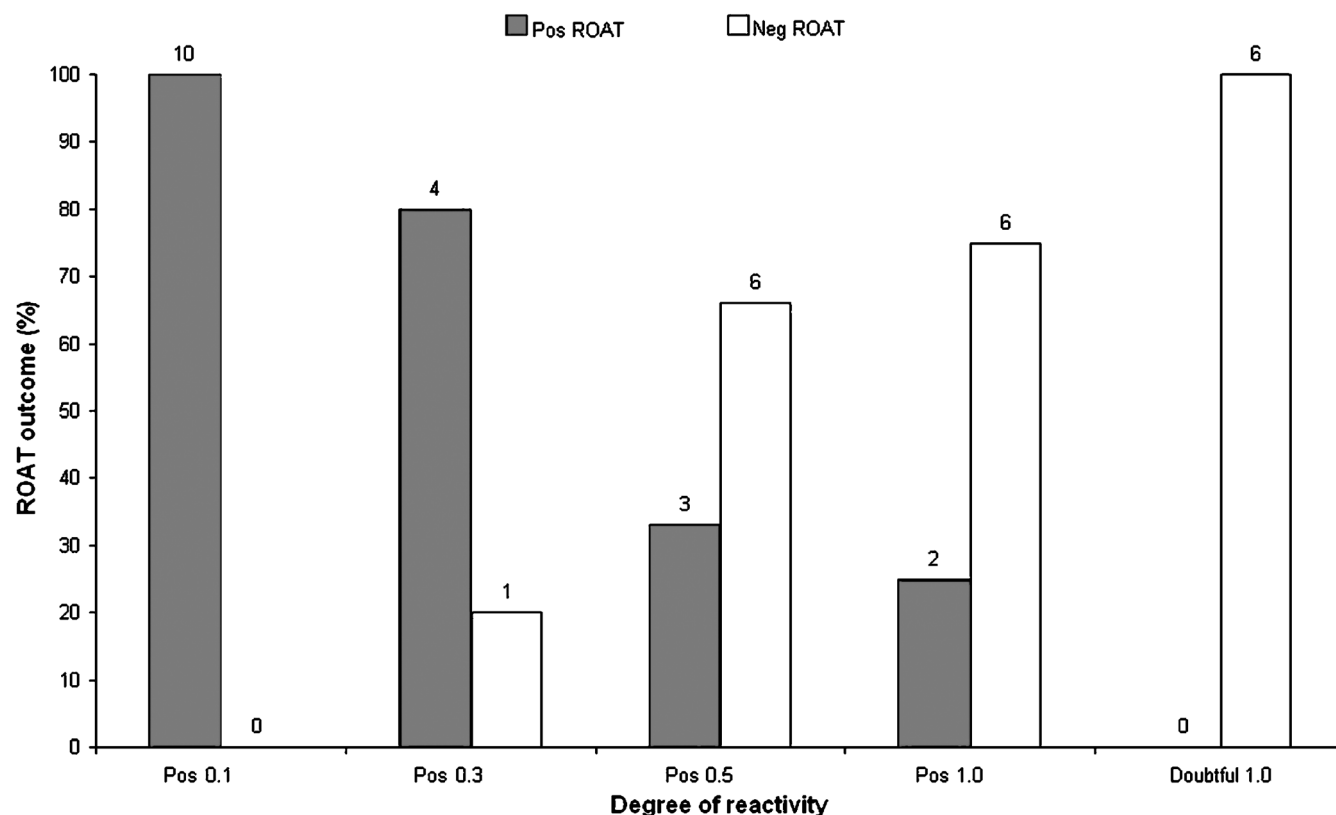


Figure 2. Distribution of positive and negative repeated open application test (ROAT) responses in numbers for the respective patch-test reactivity levels among 32 patients who had allergic reactions and 6 patients who had doubtful reactions to methyldibromoglutaronitrile. (Neg = negative; Pos = positive)

individuals but not in controls.³¹ We recently published a report of a case of allergic contact dermatitis from MDBGN in a liquid soap. This patient only reacted to MDBGN at 1.0% and not at concentrations of 0.5% and less when patch-tested (results read on day 3).³²

Conclusions

This study demonstrates that patch testing with MDBGN at 0.3% and 0.1% will miss clinically relevant patch-test reactions to MDBGN. With the present knowledge of rates of contact allergy to MDBGN and clinical relevance, patients with positive patch-test reactions should avoid exposure to MDBGN. Furthermore, the circumstances of high and increasing rates of European contact allergy to MDBGN, the reports on anecdotal cases of allergic contact dermatitis from MDBGN, and the ROAT results of this study and the previous study mentioned merit legislative measures to restrict or ban both non-occupational and occupational exposures to MDBGN, particularly from leave-on products, to prevent sensitization to MDBGN and elicitation of dermatitis from MDBGN in persons who are already sensitized. Since July 2004, MDBGN has been

banned in cosmetics of leave-on type in the European Community, but patients may still be exposed to MDBGN at concentrations up to 0.1% in rinse-off products.

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