

Is Quaternium-15 a Formaldehyde Releaser? Correlation Between Positive Patch Test Reactions to Formaldehyde and Quaternium-15

Ashika Odhav and Donald V. Belsito, MD

Background: The question of whether quaternium-15 is a formaldehyde releaser is controversial. Understanding this relationship is critical because of the widespread use of quaternium-15 and the need to properly advise formaldehyde-allergic individuals.

Objectives: This study aimed to look for an association between allergy to quaternium-15 and formaldehyde by correlating reactions to both and to correlate the intensity of positive patch test results to formaldehyde with reactivity to quaternium-15.

Methods: This is a retrospective analysis of 1905 patients who underwent patch testing for allergic contact dermatitis. Associations were analyzed by χ^2 testing.

Results: Of all patients, 9.5% reacted to quaternium-15, 7.2% reacted to formaldehyde, and 5.4% reacted to both ($P < 0.001$). Of 137, 86 had strong (2 or 3+) and 51 had weak (1+ or +/-) formaldehyde reactions; there was no relationship between the severity of formaldehyde reactivity and responsiveness to quaternium-15 ($P = 0.229$). Sex analysis did not change these findings.

Limitations: This study is limited by its retrospective analysis and small sample size.

Conclusions: A statistically significant relationship exists between reactivity to quaternium-15 and formaldehyde; however, the severity of the formaldehyde reaction does not predict reactivity to quaternium-15. Despite coreactivity with formaldehyde, quaternium-15 may not be a significant formaldehyde releaser. The coreactivity between quaternium-15 and formaldehyde requires further studies.

QUATERNIUM-15 is one of the most widely used topical preservatives. It is a quaternary ammonium salt that is a broad-spectrum antimicrobial agent used in cosmetic and industrial products. Shampoos and conditioners, liquid soaps, eye makeup, foundation makeup and powders, lotions and creams, shaving products, and bath gels are among those consumer products that commonly contain quaternium-15.¹ The concentration of quaternium-15 in cosmetics varies between 0.000002% and 0.2%.^{2,3} It is also found in noncosmetic products such as floor waxes and polishes, inks, textile finishing solutions, and printing paste.² Despite identical restrictions in the United States and Europe

regarding the maximum concentrations of quaternium-15 in cosmetics, the prevalence of positive patch test reactions to quaternium-15 in the United States is higher than that in Europe.⁴ The authors hypothesized that this might possibly be due to exposure patterns, overall higher concentrations of quaternium-15 in cosmetic products in the United States, and/or greater exposure to noncosmetic products in the United States containing quaternium-15. Another possible explanation is less access to patch testing in the United States resulting in higher rates of positive reactions among patients in the United States undergoing patch tests.⁵

The data in the literature are controversial as to whether quaternium-15 is a formaldehyde releaser or not. Most dermatologists have been taught that, on hydrolysis, quaternium-15 releases formaldehyde. This belief was based on several studies that detected formaldehyde in cosmetic products known to contain quaternium-15.⁶⁻⁹ However, these data are speculative because quaternium-15 was not tested neat; therefore, the source of formaldehyde cannot be definitively ascribed to quaternium-15.

More convincing data that quaternium-15 is a formaldehyde releaser come from several other studies. Using highperformance liquid chromatography, Benassi et al¹⁰ demonstrated that a 0.2% solution of quaternium-15 in water showed a steady-state level of

From the Department of Medicine (Dermatology), University of Missouri – Kansas City, MO.

Address reprint requests to Donald V. Belsito, MD, Department of Dermatology, Columbia University Medical Center, New York, NY 10032. E-mail: dvb2108@columbia.edu.

The authors have no funding or conflicts of interest to declare.

Dr Belsito is an expert panel member of the Cosmetic Ingredient Review, which recently reviewed the safety of quaternium-15 in cosmetics.

DOI: 10.1097/DER.0b013e31823d1785

© 2012 American Contact Dermatitis Society. All Rights Reserved.

300 µg/mL (300 ppm) of formaldehyde at around 60 days. Kijami et al,¹¹ also using high-performance liquid chromatography, determined that a 0.1% aqueous solution of quaternium-15 releases approximately 700 ppm of formaldehyde. Finally, using acetylacetone colorimetry, Wu et al⁶ found that a 0.2% solution of quaternium-15 liberated 1176 ppm of formaldehyde.

In contrast to these, studies by Dow Chemical Co, a manufacturer of quaternium-15, indicate that quaternium-15 may not be a formaldehyde releaser. In 1982, Dow Chemical Co tested an aqueous solution of 0.2% quaternium-15 by C-13 nuclear magnetic resonance spectroscopy. The C-13 nuclear magnetic resonance spectra were scanned on the 11th, 28th, and 29th days of storage of the neat 0.2% solution at room temperature. The results revealed there was no absorption detected in the target region in any spectra, which indicated that formaldehyde was not present in quantities greater than the estimated 70-µg/mL (70 ppm) detection limit.² Using the same technology, Dow Chemical Co analyzed a shampoo containing 0.1% quaternium-15 for the presence of formaldehyde. After the neat shampoo sample was scanned, the results indicated no formaldehyde was detected at the range limit of 60 µg/g (60 ppm).²

Given the controversial data in the literature, we undertook a retrospective study to (1) statistically correlate the incidence of positive patch test reactions to both quaternium-15 and formaldehyde and, (2) if a statistically significant correlation existed, to correlate the intensity of positive patch tests to quaternium-15 with those to formaldehyde. Our hypothesis was that, if quaternium-15 is a formaldehyde releaser, then individuals with more intense reactions to formaldehyde should be more likely to react to quaternium-15 than patients with weak or no reaction to formaldehyde.

PATIENTS AND METHODS

Between July 1, 1994, and June 30, 2010, a total of 1905 patients with suspected allergic contact dermatitis were patch tested by the senior author with the North American Contact Dermatitis Group's standard allergen series, which contained 2.0% quaternium-15 in petrolatum and 1.0% formaldehyde in water. Patients were patch tested in a standardized manner using Finn Chambers on Scanpor tape.¹² Test allergens were purchased from Chemo-technique Diagnostics AB, Malmö, Sweden (1994–2007), or from allergEAZE, SmartPractice, Calgary, AB, Canada (2008–2010).

Allergens were applied on Mondays, and patients were examined at days 2 and 4. Reactions were graded on morphology as previously defined.¹² Reactions scored as +, ++, and +++ were considered allergic, whereas reactions scored as +/- were doubtful. A 1+, 2+, or 3+ final reading was interpreted as a positive response, whereas a +/- response was considered potentially positive.

All deidentified Health Insurance Portability and Accountability Act-compliant data were entered, retrieved, and evaluated using a computer database (Access 2007; Microsoft Corp, Seattle, Wash). A χ^2 test was conducted to test whether the difference between observed and expected frequencies was statistically significant. Marginal frequencies and χ^2 tests were calculated to express the relationship of coreactivity versus cross-reactivity between formaldehyde and quaternium-15 using Research and Statistical Consult Service (University of Missouri – Kansas City, Mo).

RESULTS

As shown in Table 1, of the 1905 patients tested, 9.5% (n = 181) exhibited an allergic reaction to quaternium-15 and 7.2% (n = 137) exhibited an allergic reaction to formaldehyde. There were 5.4% (n = 102) who exhibited allergic reactions to both. Given the marginal frequencies for positive reactions to each allergen, the expected frequency of patients reacting to both was 0.7% (n = 13). The χ^2 test analysis showed that the relationship between allergies to formaldehyde and quaternium-15 was statistically significant with the observed frequencies differing from the expected frequencies ($P < 0.001$; Table 1).

The above finding held true when analyzed by sex (Tables 2 and 3). Among the 1213 females in the study, 83 (6.9%) had a reaction to formaldehyde, 115 (9.6%) had an allergic reaction to quaternium-15, and 62 (5.2%) had reactions to both (vs an expected frequency of 8 cases). Among the 692 males in the study, 54 (7.8%) had a reaction to formaldehyde, 66 (9.6%) had an allergic reaction to quaternium-15, and 40 (5.8%) had reactions to both (vs an expected frequency of 5 cases).

To determine whether the severity of the formaldehyde reaction correlated with an allergic reaction to quaternium-15, an additional χ^2 test was conducted using only patients exhibiting allergic reactions to formaldehyde. Formaldehyde-allergic patients were split into 2 levels of reactivity: weak (reactions of 1+ and +/-) and strong (2+ and 3+). A total of 137 patients tested positive for

TABLE 1. Frequency of Positive Patch Test Reactions in Patients Tested to Both Quaternium-15 and Formaldehyde*

	Formaldehyde (–)	Formaldehyde 2 or 3+	Formaldehyde 1+ or +/-	Formaldehyde 1 or 2 or 3 or 4
Quaternium-15 (–)	1675	19	16	35
Quaternium-15 2 or 3+	54	57	16	73
Quaternium-15 1 or +/-	25	10	19	29
Quaternium-15 1 or 2 or 3 or 4	79	67	35	102

*A total of 1905 patients were tested to both formaldehyde and quaternium-15. In addition to the 1891 patients delineated in this table, 7 had irritant reactions to formaldehyde and were negative to quaternium-15, 2 had irritant reactions to both patch test preparations, and 5 had irritant reactions to quaternium-15 and were negative to formaldehyde.

TABLE 2. Frequency of Positive Patch Test Reactions in Female Patients Tested to Both Quaternium-15 and Formaldehyde*

	Formaldehyde (–)	Formaldehyde 2 or 3+	Formaldehyde 1+ or +/-	Formaldehyde 1 or 2 or 3 or 4
Quaternium-15 (–)	1067	8	13	21
Quaternium-15 2 or 3+	42	31	9	40
Quaternium-15 1 or +/-	11	6	16	22
Quaternium-15 1 or 2 or 3 or 4	53	37	25	62

*A total of 1213 female patients were tested to both formaldehyde and quaternium-15. In addition to the 1203 patients delineated in this table, 4 had irritant reactions to formaldehyde and were negative to quaternium-15, 3 had irritant reactions to both patch test preparations, and 3 had irritant reactions to quaternium-15 and were negative to formaldehyde.

formaldehyde, of whom 51 were weakly and 86 strongly positive to formaldehyde. Of the formaldehyde positive patients, 35 patients tested negative to quaternium-15 and 102 were positive. The analysis revealed that there was no relationship between the severity of the formaldehyde response and the presence of a quaternium-15 allergic reaction ($r_{135} = 0.10$, $\chi^2_1 [n = 137] = 1.45$, $P = 0.229$). In order for these results to be statistically significant with a power of 80% (effect size, 0.103), a minimum of 727 patients who test positive for formaldehyde would be needed. This represents an increase of 590 patients from the 137 in the current analysis; assuming the rate of formaldehyde allergy in new patients would be the same as the current sample (ie, 7.2%), then 10,100 patients would need to have been screened. Subanalysis by sex did not change the finding of a lack of correlation between the intensity of the formaldehyde reaction and a reaction to quaternium-15.

We further analyzed our data as to whether there was a correlation between (a) the degree of formaldehyde reactivity $\geq$ that of quaternium-15 and (b) the degree of formaldehyde reactivity $>$ that of quaternium-15, regardless of the intensity of the formaldehyde reaction. Among the 102 patients displaying allergic reactions to both quaternium-15 and formaldehyde, the reactions for 45 patients (44.1%) were equally severe for both allergens, the reactions of 43 (42.2%) were more severe to quaternium-15, and the reactions of 14 (13.7%) were more severe for formaldehyde. Overall, there were 59 patients (ie, 57.8%) with reactions to formaldehyde that were equally or more severe than the reaction to quaternium-15. We assumed that a more severe reaction to one or the other happening by chance would display a 50:50 split (ie, 51 of 102 patients with a more severe reaction to formaldehyde and

51 with a greater reaction to quaternium-15). We conducted 3 χ^2 tests against the results we would expect by chance.

First, we tested whether the number of patients with equal or more severe reaction to formaldehyde was statistically different from that expected by chance (ie, does 59 significantly differ from 51); the χ^2 test revealed that it was not ($\chi^2_1 [n = 102] = 2.51$, $P = 0.113$). Second, we tested whether the number of patients with a more severe reaction to formaldehyde was different from the 50% expected by chance (ie, does 14 significantly differ from 51); the χ^2 test revealed that association was significantly less than that expected by chance ($\chi^2_1 [n = 102] = 53.69$, $P < 0.001$). For the final test, we excluded those patients with equally severe reactions to quaternium-15 and formaldehyde and tested whether the number with more severe reactions to formaldehyde differed from the number with more severe allergies to quaternium-15. In other words, among the 57 patients with more severe reactions to one or the other allergen, was one of the allergens more likely than chance to elicit a more severe reaction (ie, does 43 [quaternium-15] differ significantly from 14 [formaldehyde]). The χ^2 test revealed that quaternium-15 was significantly more likely to elicit a more severe reaction than formaldehyde ($\chi^2_1 [n = 57] = 14.75$, $P < 0.001$). Subanalysis by sex (Tables 2 and 3) had no effect on the previously mentioned findings.

DISCUSSION

Our data indicate that quaternium-15 significantly coreacts with formaldehyde. Because it is unlikely that a manufacturer would intentionally combine quaternium-15 and formaldehyde in the

TABLE 3. Frequency of Positive Patch Test Reactions in Male Patients Tested to Both Quaternium-15 and Formaldehyde*

	Formaldehyde (–)	Formaldehyde 2 or 3+	Formaldehyde 1+ or +/-	Formaldehyde 1 or 2 or 3 or 4
Quaternium-15 (–)	608	11	3	14
Quaternium-15 2 or 3+	12	26	7	33
Quaternium-15 1 or +/-	14	4	3	7
Quaternium-15 1 or 2 or 3 or 4	26	30	10	40

*A total of 692 male patients were tested to both formaldehyde and quaternium-15. In addition to the 688 patients delineated in this table, 3 had irritant reactions to formaldehyde and were negative to quaternium-15; and, 1 had an irritant reaction to quaternium-15 and was negative to formaldehyde.

same product, this relationship may be caused by (1) formaldehyde release from quaternium-15 or another ingredient in a quaternium-15 containing cosmetic, (2) formaldehyde contamination of an ingredient other than quaternium-15 in the quaternium-15 product that induced sensitization, or (3) formaldehyde contamination of quaternium-15 itself. The possibility of formaldehyde contamination of quaternium-15 is unlikely given its reported manufacturing process: “quaternary compounds are produced by reacting hexamine with the appropriate halocarbon in a non-aqueous solvent at room temperature.”²

To assess the potential that at least some of the quaternium-15 reactions were due to formaldehyde release, the intensity of the formaldehyde reaction was correlated with the frequency of quaternium-15 patch results. As demonstrated, there was no significant relationship in the severity of formaldehyde response and the presence of quaternium-15 reaction. This suggests that quaternium-15 may not be a significant formaldehyde releaser. However, as noted, if the sample size was increased to 10,100 patients, and the results remained the same, a statistically significant relationship would have been obtained.

It is important to remember that most, but not all, individuals allergic to quaternium-15 were also allergic to formaldehyde (102/181; 56.4%). Thus, many individuals can be allergic to quaternium-15 solely based on its molecular structure. Others may be allergic to both formaldehyde and quaternium-15, irrespective of formaldehyde release by quaternium-15. Therefore, we further analyzed our data as to whether there was a correlation between (a) the degree of formaldehyde reactivity $\geq$ that of quaternium-15 and (b) the degree of formaldehyde reactivity $>$ that of quaternium-15. The analysis for formaldehyde reactivity $\geq$ quaternium-15 reactivity did not show a significant correlation, whereas that for formaldehyde reactivity $>$ quaternium-15 reactivity was significantly less than expected ($P < 0.001$). Overall, patients with reactions to both allergens tend to have more severe reactions to quaternium-15. Although one could hypothesize that this results from a combination of reactivity to quaternium-15 itself plus reactivity to quaternium-15's release of formaldehyde, the statistical power of the association (as well as the 79 [43.6%] of patients who reacted only to quaternium-15) would argue against this.

In a recent study, Hauksson et al¹³ suggested that formaldehyde 2% in water should be used in patch testing rather than 1%, which was tested in our study. They argued that some clinically relevant formaldehyde allergy can be missed when testing at 1%. However, given our data that, when coreactivity between quaternium-15 and formaldehyde was observed, quaternium-15 was more likely than formaldehyde to induce severe reactions, it is unlikely that more weak reactions and/or enhanced reactivity of the existing responses to formaldehyde (induced by a higher test concentration) would significantly change our results.

As shown in Figure 1, the molecular structures of quaternium-15 and formaldehyde are very different. Because the preservatives are not structurally related, cross-reactions based on molecular structure are unlikely. According to Daniel C. Liebler, PhD, of

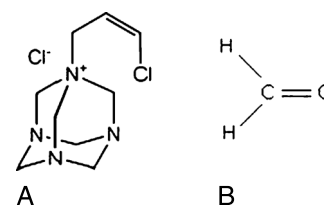


Figure 1. Molecular structures of quaternium-15 (A) and formaldehyde (B).

Vanderbilt University, “The structure [of quaternium-15], [does not provide for] a straightforward way to get to formaldehyde—certainly not through any solution chemistry. [While] oxidative metabolism of the methylene carbons in the heterocyclic rings [could] release formaldehyde, [it is not likely] that the required double-*N*-dealkylation reactions would yield formaldehyde very efficiently. [What] stands out in the structure [of quaternium-15] is the vinyl chloride ‘flag’ on the top. Oxidation of the double bond with elimination of the chloride could yield an aldehyde attached to the rest of the molecule.” Whether this aldehyde would be formaldehyde and whether this could be effectively released from the rest of the molecule is unclear. Therefore, the possibility that quaternium-15 could release formaldehyde under the correct conditions remains open. What is clear is that, based on the method of manufacturing, contamination of quaternium-15 by formaldehyde is not likely.

Adding to the confusion regarding formaldehyde release by quaternium-15 are contradictory studies specifically designed to evaluate release of quaternium-15 by formaldehyde. Although studies by Dow Chemical Co would indicate that quaternium-15 does not release formaldehyde,² 3 other independent studies,^{6,10,11} including 2 studies^{10,11} that used the Dow Chemical Co material, clearly showed levels of formaldehyde in neat preparations of 0.2% quaternium-15 ranging from 300 to 1176 ppm of formaldehyde.

In conclusion, our findings are in agreement with previously reported data^{9,14–21} showing statistically significant patch test coreactivity between quaternium-15 and formaldehyde. Whereas de Groot et al²² found that, in women, but not in men, stronger reactions to formaldehyde were associated with more positive quaternium-15 patch tests, our data do not substantiate this sex-related difference. Overall, our data and those for men from de Groot et al²² do not support release of formaldehyde by quaternium-15 as a potential cause for their coreactivity. Nonetheless, pending further studies to elucidate whether, under the proper conditions of pH, temperature, and other yet-to-be-determined factors, quaternium-15 can release significant levels of formaldehyde in cosmetic formulations, individuals allergic to formaldehyde should be cautioned regarding exposure to quaternium-15.

ACKNOWLEDGMENTS

The authors thank the following: Daniel C. Liebler, PhD, (Director, Jim Ayers Institute for Precancer Detection and Diagnosis, Ingram Professor of Cancer Research, Professor of Biochemistry,

Pharmacology and Biomedical Informatics, Vanderbilt University School of Medicine), for his insight on the chemical structures and metabolism of quaternium-15 and Aaron Bonham (statistician at Research and Statistical Consult Service at University of Missouri Kansas City) for statistical analysis of the data.

REFERENCES

1. Elder RL, ed. Final report on the safety assessment of quaternium-15. *J Environ Pathol Toxicol* 1986;5:61–101.
2. Bergfeld WF, Belsito DV, Klaassen CD, et al. Amended safety assessment of quaternium-15 as used in cosmetics. *Int J Toxicol* 2010;29(Suppl 3):98S–114S.
3. Regulation (EC) No. 1223/2009 of the European Parliament and of the Council of 30 November 2009 on cosmetic products. Available at: <http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2009:342:0059:0209:EN:PDF>. Accessed September 3, 2010.
4. de Groot AC, Veenstra M. Formaldehyde-releasers in cosmetics in the USA and in Europe. *Contact Dermatitis* 2010;62:221–224.
5. Statham B, Mughal A, Bodger O. The relationship between the proportion of the population tested annually and the rate of patients with positive patch test reactions. *Contact Dermatitis* 2011;64:54–57.
6. Wu P, Chang C, Chou S. Determination of formaldehyde in cosmetics by HPLC method and acetylacetone method. *J Food Drug Anal* 2003;11:8–15.
7. Scott CR, Wolf PA. The antibacterial activity of a series of quaternaries prepared from hexamethylene tetraamine and haloalkylcarbons. *Appl Microbiol* 1962;10:211–216.
8. Jordan WP, Sherman WT, King SE. Threshold responses in formaldehyde-sensitive individuals. *J Am Acad Dermatol* 1979;1:44–48.
9. de Groot AC, White IR, Flyvholm M, et al. Formaldehyde-releasers in cosmetics: relationship to formaldehyde contact allergy. Part 2. *Contact Dermatitis* 2010;62:18–31.
10. Benassi CA, Semenzato A, Zaccaria F, et al. High performance liquid chromatographic determination of free formaldehyde in cosmetics preserved with Dowicil 200. *J Chromatogr* 1990;502:193–200.
11. Kijami K, Takeda M, Okaya Y, et al. A study on release of formaldehyde from its donor type preservatives. *Anal Sci* 1991;7(Suppl):913–916.
12. Storrs FJ, Rosenthal LE, Adams RM, et al. Prevalence and relevance of allergic reactions in patients patch tested in North America—1984 to 1985. *J Am Acad Dermatol* 1989;20(6):1038–1045.
13. Hauksson I, Pontén A, Gruvberger B, et al. Clinically relevant contact allergy to formaldehyde may be missed by testing with formaldehyde 1.0%. *Br J Dermatol* 2010;164:568–572.
14. Lundov M, Johansen J, Carlsen B, et al. Formaldehyde exposure and patterns of concomitant contact allergy to formaldehyde and formaldehyde-releasers. *Contact Dermatitis* 2010;63:31–36.
15. Aalto-Korte K, Kuuliala O, Suuronen K, et al. Occupational contact allergy to formaldehyde and formaldehyde releasers. *Contact Dermatitis* 2008;59:280–289.
16. Statham BN, Smith EV, Bodger OG, et al. Concomitant contact allergy to methylchloroisothiazolinone/methylisothiazolinone and formaldehyde-releasing preservatives. *Contact Dermatitis* 2010;62:56–57.
17. Jacobs M-C, White IR, Rycroft RJG, et al. Patch testing with preservatives at St John's from 1982 to 1993. *Contact Dermatitis* 1995;33:247–254.
18. Parker LU, Taylor JS. A 5 year study of contact allergy to quaternium-15. *Am J Contact Derm* 1991;2:231–234.
19. Fisher AA. Cosmetic dermatitis. Part II. Reactions to some commonly used preservatives. *Cutis* 1980;26:136.
20. Ford GP, Beck MH. Reactions to quaternium 15, Bronopol and Germall 115 in a standard series. *Contact Dermatitis* 1986;14:271–274.
21. Emeis D, de Groot AC, Brinkmann J. Determination of formaldehyde in formaldehyde-releaser patch test preparations. *Contact Dermatitis* 2010;63:57–62.
22. de Groot AC, Blok J, Coenraads P-J. Relationship between formaldehyde and quaternium-15 contact allergy. Influence of strength of patch test reactions. *Contact Dermatitis* 2010;63:187–191.