

Cosmetics and herbal remedies with Compositae plant extracts – are they tolerated by Compositae-allergic patients?

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Background: Compositae-sensitive patients are routinely warned against topical use of Compositae-containing cosmetics and herbal remedies. However, the risk of elicitation of dermatitis in presensitized persons is unknown.

Objectives: The main aim of this study was to assess the significance of direct plant allergen contact via Compositae-derived cosmetics and herbal remedies in Compositae-allergic patients with special reference to arnica (*Arnica montana*) and German chamomile (*Chamomilla recutita*).

Methods: 8 of 12 chamomile-sensitive patients tested positive to chamomile-containing preparations, including tea, creams, ointments, and oil. 5 of 6 arnica-sensitive persons tested positive to arnica-based products.

Results: When the group was patch tested with cosmetic and/or herbal product ingredients, plant allergens elicited positive reactions most frequently, but fragrances, emulsifiers, and preservatives tested positive as well. Plant allergens were mainly derived from Compositae, but avocado oil, and *Hamamelis virginiana* tincture were unexpectedly detected as sensitizers too. Chemical analyses indicated that the Compositae allergens were both sesquiterpene lactones and other naturally occurring compounds.

Conclusion: In conclusion, Compositae-allergic persons should be warned against topical use of Compositae-containing products, not only because of the plant allergens, but also because of allergenic cream constituents that may cause reactions in the group of patients who have multiple contact allergies beside the Compositae allergy.

Key words: allergic contact dermatitis; *Arnica montana*; avocado oil; *Chamomilla recutita*; cosmetics; *Hamamelis virginiana*; herbal medicine; patch testing; *Persea americana* Mill; *Persea gratissima*.
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The abundance of Compositae-containing cosmetics and topical remedies is considered a risk factor for Compositae-sensitive patients. The most widely used Compositae plants in this connection are probably arnica (*Arnica montana* L.) and 2 species of chamomile, German chamomile (*Chamomilla recutita* (L.) Rauschert) and Roman chamomile (*Chamaemelum nobile* (L.) All. = *Anthemis nobilis* L.). On the basis of scarce epidemiological data, sensitization has occurred relatively frequently with arnica tincture, at least formerly, and rarely with the widely used

German chamomile, although sensitization has been described with a variety of chamomile preparations, including chamomile oil, chamomile tea, and sesquiterpene lactone-free chamomile cultivars (1). However, the magnitude of the risk of sensitization, or elicitation of dermatitis in previously Compositae-sensitized subjects, is unknown.

The aims of this study were twofold: (i) to assess the significance of direct allergen contact via Compositae-containing topical herbal remedies and cosmetics in Compositae-sensitive patients with special reference to arnica and German

chamomile and, if plant allergy was shown, (ii) to detect and characterize possible allergens/allergenic fractions from the plant extracts.

Patient Population, Materials and Methods

Adult, non-pregnant persons who had previously tested positive (at least 2+) to short ether extracts of German chamomile 2.5% petrolatum (pet.), and/or arnica 0.5% pet. at our department were invited to participate.

Chamomile-sensitive persons were re-tested with German chamomile to verify the allergy (unless they had been tested within the last 6 months) and additionally with Roman chamomile and chamomile-derived or chamomile-containing products as shown in Table 1. The teas and the commercial preparations, which were all stay-on products, were tested 'as is', whereas the blue chamomile flower oil, derived from German chamomile and also called German chamomile oil, chamomile blue oil, Hungarian chamomile flower oil etc., was tested 4% and 1% in pet.

The chamomile tea was made by adding 1 tea bag or 1 top teaspoon of dried flowers to about 200 ml of boiling water for 8–10 min.

Arnica-sensitive patients were similarly tested with the extracts and products shown in Table 2. 2 arnica extracts were used because the patients had been tested with extracts from different sources in the previous years. Some preparations contained both chamomile and arnica and were accordingly tested in both groups of patients.

Products eliciting positive patch test reactions in at least 3 subjects were studied further. In case of positive reactions to commercial preparations, the patients were patch tested with allergenic ingredients of the products in question; furthermore, the raw *Compositae* plant material, provided by the different manufacturers, were

screened for sesquiterpene lactones (SLs) and other potential allergens at the Department of Food Science, University of Aarhus, Research Centre Aarslev, Denmark.

All patch test materials were applied to the back for 2 days using Finn Chambers[®] (Epitest Ltd. Oy, Tuusula, Finland) on Scanpor[®] tape (Alpharma AS, Vennesla, Norway) and readings performed on D3 and possibly D7 according to International Contact Dermatitis Research Group (ICDRG) recommendations (2). The patients were tested between October and May, in a 2-year period from 2003 to 2005, to avoid possible summer exacerbation affecting the test results. The study was approved by the local ethics committee (file no. VF 20030001).

Chemical analysis of ingredients

Allergenic ingredients (tinctures, infusions, CO₂-extract and oil) of various products were analysed without further purification or up-concentration by gas chromatography-mass spectrometry (GC-MS) for potential contact allergens with special focus on their content of SLs. GC-MS analysis were performed on a Thermo Finnigan Trace DSQ single quadrupole mass spectrometer operated at 70 eV and directly coupled to a Trace gas chromatography Ultra gas chromatograph equipped with a split/splitless PTV injector (split flow 10 ml/min, 200°C) and a Zebron capillary column (30 m × 0.25 mm i.d., film thickness df = 0.25 µm, ZB-1, Phenomenex, Torrance, CA, USA). Helium was the carrier gas at a constant column head pressure of 10 psi. The temperature of the column and transfer line was 200°C. GC-MS was performed on 2 µl samples. Compounds were separated by the following oven temperature gradient: 80°C for 1 min, programmed to 300°C at 10°C/min followed by constant temperature for

Table 1. Chamomile-containing patch test material

Product	Manufacturer
Extract of German chamomile 2.5% pet.	Trolab [®] Hermal, Reinbek, Germany
Extract of Roman chamomile 1.0% pet.	Chemotechnique [®] , Malmö, Sweden
Tea from dried German chamomile flowers.	Natur-Drogeriet A/S, Hørning, Denmark
Tea from organic Roman chamomile (tea bags)	Fredsted The A/S, Hvidovre, Denmark
Avène Soothing Eye Contour Cream	Pierre Fabre, Laboratoires Dermatologiques Avène, Paris, France
A.Vogel Bioforce Crème -7 Herb Cream	Bioforce AG, Roggwil, Switzerland
St. Ives Citrus & Ginseng Energising Hand & Body Lotion	Cederroth, Upplands Väsby, Sweden
Dr Scheller Chamomile Ointment	Dr Scheller Cosmetics AG, Eislingen, Germany
Dr Scheller Camomile Herbal Blossom Hand lotion	Apotheker Dr Scheller, Göppingen, Germany
Mellisa Herbal Ointment	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark
Mellisa Day Cream with marigold, German chamomile, and arnica	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark
Mellisa Arnica Ointment	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark
Blue chamomile flower oil 4.0% pet.	Nordlys Centret, Silkeborg, Denmark
Blue chamomile flower oil 1.0% pet.	

pet., petrolatum.

Table 2. Arnica-containing patch test materials

Product	Manufacturer
Extract of arnica 0.5% pet.	Trolab [®] , Hermal, Reinbek, Germany
Extract of arnica 0.5% pet.	B. M. Hausen, Hamburg, Germany
Dr Scheller Arnica Herbal Blossom Hand Creme	Apotheker Dr Scheller, Göppingen, Germany
Weleda Massage Oil with arnica	Weleda, Schwäb. Gmünd., Germany
A. Vogel Bioforce Crème -7 Herb Cream	Bioforce AG, Roggwil, Switzerland
Mellisa Arnica Ointment	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark
Mellisa Day Cream with marigold, German chamomile, and arnica	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark
Mellisa Face Gel	Mellisa [®] Naturkosmetik ApS, Hørsholm, Denmark

pet., petrolatum.

20 min. The mass spectrometer was operated in full scan mode over a mass range from 39 to 600 amu (1 scan/s). SLs and other potential allergens were identified by comparing their mass spectra with literature values and/or by the use of authentic standards.

Results

Chamomile-containing products

22 patients with chamomile allergy were invited and 14 responded and were patch tested with the materials shown in Table 1. However, patient number (no.) 9, who was strongly allergic to methyl-dibromo glutaronitrile (MDBGN), was not tested

with Mellisa Day Cream, which contained this preservative (Table 3). 2 subjects, 1 of whom had just returned from a holiday and was very suntanned, had to be withdrawn from the study because of doubtful positive/negative reactions to the plant extracts (patients nos. 13 and 14, Table 3). 8 of the remaining 12 patients had 1 or more positive reactions to chamomile-containing preparations: 3 tested positive to chamomile tea, 6 to chamomile creams/ointments and 1 to chamomile oil (Table 3). The mean age of the 8 persons with positive reactions was 47 years compared with 63 years among the 4 persons with negative reactions. Table 4 shows the distribution of other contact allergies in the different subgroups of chamomile-sensitive subjects, based on the results of the latest patch testing with the standard series and selected allergens.

Arnica-containing products

Among 14 arnica-sensitive persons invited, 7 responded and were patch tested with the products shown in Table 2. Again 1 patient had to be withdrawn from further studies because of negative reactions to both arnica extracts (patient no. 21, Table 5). 5 of the other 6 patients had at least 1 positive reaction to arnica-based products (Table 5). The mean age of these 5 persons was 61 years, and the distribution of other contact allergies in arnica-sensitive patients is shown in Table 6.

The overall prevalence of positive reactions to chamomile- and/or arnica-containing preparations was 72% (13/18).

Table 3. Distribution of patch test reactions to chamomile-containing preparations in chamomile-sensitive persons. The readings shown are the maximum reactions on D3 or 7

Preparation	Patient number													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
German chamomile 2.5% pet.	NT	++	NT	NT	NT	+	++	+++	+++	++	+	++	0 F	—
Roman chamomile 1% pet.	0 F	++	0	—	—	—	—	++	+ F	—	—	0 F	—	—
Chamomile tea	0	0 F	0	0 F	—	—	—	++	—	—	+ F	—	0 F	—
Roman chamomile tea (tea bags)	+	0 F	0	—	—	—	—	++	—	—	+ F	0	—	—
Avène Eye Cream	—	—	—	—	—	—	—	—	—	—	—	—	—	—
St. Ives	0	0 F	0	0	—	—	0	—	0 F	0	—	0	—	—
Body Lotion	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Bioforce Crème	—	0	+	0 F	—	—	—	0 F	+ F	—	0 F	—	—	—
Dr Scheller	+ F	0	+	0 F	—	—	—	0 F	+ F	—	—	+	—	—
Chamomile Ointment	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Dr Scheller	+ F	—	+	0 F	—	—	0 F	++	+ F	—	—	—	—	0
Camomile Hand lotion	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Mellisa Herbal Ointment	—	—	0	—	—	—	—	—	—	—	—	—	—	—
Mellisa Day Cream	—	—	+	—	—	—	++	—	NT	0 F	—	—	—	—
Mellisa Arnica Ointment	—	—	0 F	—	—	—	—	—	—	—	—	—	—	—
Blue chamomile flower oil 4% and 1% pet.	—/—	—/—	0/—	++/++	—/—	—/—	—/—	—/—	—/—	—/—	—/—	—/—	—/—	—/—

F, follicular reaction; NT, not tested; 0, doubtful positive reaction; —, negative reaction; pet., petrolatum.

Table 4. Distribution of number of other contact allergies in 12 chamomile-sensitive patients

	Reaction to chamomile-containing products	
	Positive	Negative
Sex	7 F, 1 M	3 F, 1 M
Mean age	47 years	63 years
Other contact allergies		
0	3	0
1–2	1	1
3 or more	4	3

F, female; M, male.

Testing with ingredients

Altogether, the chamomile teas, 1 arnica- and 2 chamomile-containing products and 2 arnica- and chamomile-based preparations elicited positive reactions in at least 3 persons each and were selected for further studies. The 5 commercial products tested positive in 9 persons (patients nos. 1, 3, 7, 8, 9, 12, 15, 17, and 18).

Bioforce 7 Herb Cream contained, among other ingredients, tinctures of different plants, including chamomile and arnica. The 3 persons testing positive to the cream 'as is' (patient nos. 3, 9, and 18, Tables 3 and 5) were patch tested to another batch of the cream and the corresponding ingredients, including a mixture of all tinctures, but excluding the allegedly allergenic individual tinctures of chamomile and arnica, to rule out concomitant sensitization to other compounds. 2 subjects (patients nos. 9 and 18) tested positive to the cream again and to the mixture of all tinctures (including chamomile and arnica), but while patient no. 18 was negative to the individual tinctures, patient no. 9 had a positive reaction to *Hamamelis virginiana* (witch hazel) tincture (Table 7).

Concerning Dr Scheller Chamomile Ointment, the manufacturer provided us with the most important allergenic compounds (Table 7). It was

Table 6. Distribution of number of other contact allergies in 6 arnica-sensitive patients

	Reactions to arnica-containing products	
	Positive	Negative
Sex	3 F, 2 M	1 F
Mean age	61 years	52 years
Other contact allergies		
0	0	0
1–2	0	0
3 or more	5	1

F, female; M, male.

decided, however, not to test with the oleoresin-containing chamomile concrete because all patients were positive to the oleoresinous chamomile extract.

Similarly, we received potential allergenic emulsifiers, preservatives, fragrances, and chamomile concrete used in Dr Scheller Camomile Herbal Blossom Hand lotion (Table 7). The plant oleoresin was excluded from the patch test material for the above-mentioned reason. On the other hand, we added the following allergens listed on the ingredients label: cetyl alcohol, dexpanthenol, tocopherol acetate, Euxyl[®] K400 (a mixture of phenoxyethanol and MDBGN) as well as pure phenoxyethanol (tested in 2 patients). Only 1 person tested positive, and she was allergic to both MDBGN and phenoxyethanol (Tables 7 and 8).

The formula of the Mellisa Day Cream was changed between the first and the second period of testing, and the most important difference was the removal of MDBGN because of a growing number of reports of allergy to this preservative. The 3 subjects with positive reactions to the old formula (patients nos. 3, 7, and 15, Tables 3 and 5) were tested with the new cream as well as the ingredients of the previous formula except glyceryl stearate SE, *Citrus limonium* and *Yucca vera*. 2 of 3 persons tested positive to various ingredients,

Table 5. Distribution of patch test reactions to arnica-containing preparations in arnica-sensitive persons on D3

Preparation	Patient number						
	15	16	17	18	19	20	21
Arnica 0.5% pet. (Trolab [®])	NT	++	++	++	+++	–	–
Arnica 0.5% pet. (Hausen)	NT	++	++	+++	+++	++	0
Dr Scheller	+	0 F	+	++	0 F	0 F	–
Arnica Hand Cream							
Weleda Massage	–	0 F	0	0	++	0 F	–
Oil with Arnica							
Bioforce Crème	0 F	–	–	+ F	–	0 F	–
Mellisa Arnica Ointment	–	–	0	0	–	0 F	–
Mellisa Day Cream	++	0 F	0 F	–	0 F	0 F	–
Mellisa Face Gel	++	0 F	–	0	0	+	–

F, follicular reaction; NT, not tested; 0, doubtful positive reaction; –, negative reaction; pet., petrolatum.

Table 7. Results of testing with ingredients of chamomile- and/or arnica-containing products in chamomile-sensitive patients and arnica-sensitive patients

Product (ingredients tested)	Nos. of patients tested	Nos. positive
Chamomile-sensitive patients		
Bioforce 7 Herb Cream ('all' ingredients except chamomile and arnica tinctures)	2 (pts.nos.3 and 9)	1 (pt.no.9)
Dr Scheller Chamomile Ointment (wool alcohols, emulsifiers, preservatives, but not chamomile extract)	4 (pts.nos.1,3,9, and 12)	0
Dr Scheller Chamomile Herbal Blossom Hand lotion (emulsifiers, preservatives, fragrance, vitamins, cetyl alcohol, but not chamomile extract)	4 (pts. nos. 1, 3, 8, and 9)	1 (pt.no.9)
Mellisa Day Cream with marigold, chamomile and arnica ('all' ingredients except glyceryl stearate SE, <i>Citrus limonium</i> , <i>Yucca vera</i> , but including aqueous extracts of chamomile, arnica and marigold)	2 (pts. nos. 3 and 7)	1 (pt.no.7)
Arnica-sensitive patients		
Bioforce 7 Herb Cream	1 (pt.no.18)	1
Mellisa Day Cream with marigold, chamomile, and arnica	1 (pt.no.15)	1
Dr Scheller Arnica Herbal Blossom Hand Creme ('all' ingredients except glycine soya, dimethicone, isobutyl paraben)	3 (pts.nos.15,17, and 18)	3 (pts.nos.15,17, and 18)

pts., patients; nos., numbers.

but all 3 subjects were negative/doubtful positive to the new formula.

Concerning the Dr Scheller Arnica Herbal Blossom Hand Creme, the 3 subjects with positive reactions to the cream as is were tested with the majority of ingredients, including an extract of arnica and sunflower (*Helianthus annuus* L.) and an aqueous-alcoholic distillate of *H. virginiana*, but excluding glycine soya, dimethicone, and isobutyl paraben. The patients were tested with sodium dihydroxycetyl phosphate (SDP) 5% pet. as well as the commercial emulsifier preparation containing SDP (trade name Dragophos S 2/9 18501) 1% pet. 2 persons tested doubtful positive to Dragophos S 2/9 18501: 1 of these was also doubtful positive to SDP, while the other tested

positive. Conversely, the third person was positive to SDP and negative to Dragophos S 2/9 18501.

Tables 8 and 9 depict the distribution of reactions to ingredients, Compositae screening mixes, and other contact allergies in the 9 persons participating in this part of the study. Positive reactions to plant constituents were recorded in 5 persons. Furthermore, the negative reactions to ingredients in patients nos. 1, 8, and 12 may be indirect evidence of plant allergy because testing with the chamomile extract of 2 products was left out. Patient no. 3 had positive patch test reactions to several products (Table 3), but tested negative to all ingredients (Table 7), and the results were interpreted as false positive reactions because a flare of atopic dermatitis on his back.

Table 8. Distribution of reactions to ingredients of chamomile-containing products, Compositae screening mixes, and other contact allergens in chamomile-sensitive subjects

Patient number	Positive to ingredients	SL mix/Compositae mix 6% pet.	Other contact allergies
1	None	+++ / +++	None
3	None	+ / NT	Potassium dichromate
7	Avocado oil as is	++ / ++	Nickel sulfate, cobalt chloride, MCI/MI, Quaternium-15, formaldehyde, limonene
8	None	+++ / +++	None ^a
9	Mixture of tinctures 10% eth., <i>Hamamelis</i> tincture 10% eth., phenoxyethanol,-2	++ / ++	Nickel sulfate, FM I, FM II, <i>Myroxylon pereirae</i> resin, ethylenediamine, MCI/MI, Euxyl [®] K400, MDBGN, p-t-butylcatechol, Disperse Blue 106, oxidized linalool
12	None	++ / +++	Nickel sulfate, iodopropynyl butylcarbamate, tomato oleoresin

eth, ethanol; FM I, fragrance mix I (mixture of cinnamyl alcohol, cinnamic aldehyde, eugenol, alpha-amyl cinnamaldehyde, hydroxycitronellal, geraniol, isoeugenol and oak moss absolute); FM II, fragrance mix II (mixture of citral, citronellol, farnesol, coumarin, alpha-hexyl cinnamaldehyde, hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyr[®])); MCI/MI, methylchloroisothiazolinone/methylisothiazolinone; MDBGN, methylidibromo glutaronitrile; *Myroxylon pereirae* resin, balsam of Peru; NT, not tested with Compositae mix, but positive to individual extracts of Compositae mix; pet., petrolatum; SL mix, sesquiterpene lactone 0.1% pet.

^aPatch testing with the standard series was negative in 1997, but positive to several allergens in 2005.

Table 9. Distribution of reactions to ingredients of arnica-containing products, Compositae screening mixes, and other contact allergens in arnica-sensitive subjects

Patient number	Positive to ingredients	SL mix/Compositae mix 6% pet.	Other contact allergies
15	Extract of marigold 30% aq., extract of arnica 30% aq., MDBGN, avocado oil 'as is' glycerine 10% aq. ^a , SDP	Negative/++	Potassium dichromate, cobalt chloride, <i>p</i> -t-butylphenol formaldehyde resin, FM I, MCI/MI, Euxyl [®] K400, MDBGN, oxidized linalool
17	Arnica/sunflower extract 1% pet., fragrance 5% and 1% pet.	Negative/++	Nickel sulfate, potassium dichromate, FM I, colophonium, PPD
18	Mixture of tinctures 10% eth., arnica/sunflower extract 1% pet., fragrance 5% and 1% pet., SDP	+++ /+++	FM I, thiuram mixture, tomato oleoresin

aq., aqueous; eth., ethanol; FM I, fragrance mix I (mixture of cinnamyl alcohol, cinnamic aldehyde, eugenol, alpha-amyl cinnamaldehyde, hydroxycitronellal, geraniol, isoeugenol, oak moss absolute); MCI/MI, methylchloroisothiazolinone/methylisothiazolinone; MDBGN, methylidibromo glutaronitrile; pet., petrolatum; PPD, *para*-phenylenediamine; SDP, sodium dihydroxycetyl phosphate; SL mix, sesquiterpene lactone mix 0.1% pet.

^aIngredient of the new formula to which the patient tested doubtful positive.

Table 10. List of non-standardized ingredients of Compositae-containing products eliciting positive patch test reactions in Compositae-sensitive individuals

Mixture of tinctures	10% eth.
Tincture of <i>Hamamelis virg.</i>	10% eth.
Extract of marigold	30% aq.
Extract of arnica	30% aq.
Avocado oil 'as is'	
Glycerine	10% aq.
Arnica/sunflower extract	1% pet.
Fragrance	5% and 1% pet.

aq., aqueous; eth., ethanol; pet., petrolatum.

Table 10 shows the non-standardized allergens of the 5 products: all were negative in 10 controls.

Chemical investigations

Arnica preparations are known for their content of pseudoguaianolide SLs (3, 4). The GC-MS analysis of arnica-containing ingredients (CO₂-extract, tincture and infusion) showed the presence of several pseudoguaianolide SLs between 19 and 22 min in the GC-MS chromatograms, including helenalin and 11 α ,13-dihydrohelenalin as well as short chain esters of 11 α ,13-dihydrohelenalin (6-*O*-acetyl-, 6-*O*-isovaleryl-, 6-*O*-isobutyryl- and 6-*O*-methacryloyl-11 α ,13-dihydrohelenalin). All detected SLs produced peaks at *m/z* 95, 96, 122, 123, 124, and 191, which are characteristic fragments of pseudoguaianolides bearing oxygen functions at the 4, 6, and 8 positions as in the case of helenalin and dihydrohelenalin and their ester derivatives (5–7). The presence of helenalin and 11 α ,13-dihydrohelenalin was verified by authentic standards. The short chain esters of dihydrohelenalin produced molecular ions ([M]⁺), at 306, 332, 334, and 348, respectively, corresponding to the acetyl, methacryloyl, isobutyryl, and isovaleryl

esters of 11 α ,13-dihydrohelenalin, respectively. In addition, these dihydrohelenalin ester derivatives produced characteristic fragments at *m/z* 264, 263, 246, and 231, which further supported the presence of these SLs in the arnica-containing ingredients (6). The content of SLs in the CO₂-extract was found to be much higher compared with the tincture and the infusion where SLs only appeared in minute or trace amounts.

The GC-MS analysis of blue chamomile flower oil showed the presence of a large number of sesquiterpenes, including chamazulene, (–)- α -bisabolol, (–)- α -bisabololoxide A and (–)- α -bisabololoxide B as the main constituents together with diacetylenic C₁₃-spiroacetal enol ethers, which is in accordance with previous investigations of the essential oil of chamomile (8). The compounds were identified by the use of authentic standards and/or by comparison with mass spectra in the National Institute of Standard Technology (NIST)/ Environmental Protection Agency (EPA)/ National Institutes for Health (NIH) Mass Spectral Library (NIST 05) database of the GC-MS. The investigation of the tincture and the infusion of German chamomile showed in addition to bisabololoxides and polyacetylenes also the presence of 7-methoxycoumarin (herniarin). The investigation of the different types of chamomile preparations did, however, not show the presence of SLs. The same was true for the investigation of the tincture and the infusion of marigold.

Discussion

The prevalence of positive reactions to products 'as is' was high and varied in these selected groups of patients. The majority of reactions were elicited by complex commercial preparations such as

creams, ointments, and lotions, and fewer by chamomile tea and oil: among 11 commercial stay-on products, 7 elicited positive reactions (Tables 3 and 5). It is hardly surprising that a mixture of potential allergenic constituents would elicit positive patch test reactions in broadly sensitized patients who comprised the majority of persons participating in this study.

In several cases, the patch test reactions to the creams could be attributed to contact allergies that the patients had been diagnosed with previously. Thus, patients nos. 9 and 15 had positive reactions to both Euxyl® K400 and MDBGN of the standard series: patient no. 9 was not tested with a cream preserved with MDBGN, but turned out to be positive to phenoxyethanol as well, which was used as a preservative in another product (Table 8). Patient no. 15 was unintendedly tested with the MDBGN-containing product and was positive to this preservative and other ingredients (Table 9). Likewise, patients nos. 17 and 18, with strongly positive fragrance mix reactions attributable to oak moss, tested positive to the fragrance of a product containing *Evernia prunastri*-extract. Furthermore, 2 patients tested positive to SDP, which is a surfactant in the emulsifier Dragophos S2/9 18501. SDP has been reported as a cosmetic allergen, but the test concentration of 5% pet. is weakly irritant (9), and only 1 of the 2 persons tested doubtful positive to Dragophos 1% pet., indicating a weak sensitization.

By far the largest group of allergens eliciting positive reactions were plant-derived materials: 5 of 9 patients tested positive, and at least 3 were positive to Compositae extracts (Tables 8 and 9). Patient no. 9 was positive to a Compositae-containing mixture of tinctures, but also tested positive to *Hamamelis* tincture, which was included in the mixture.

Patient nos. 1, 8, and 12, who were positive to 1 or 2 chamomile-containing products, tested negative to selected ingredients of both products. However, testing with the chamomile extract (chamomile concrete) was left out, and the negative reactions may thus, with reservations, be regarded as indirect evidence of Compositae reactivity. This is supported by the fact that these 3 patients had no or relatively few other contact allergies at the time of testing. Thus, the number of patients with definite or probable reactions from plant allergens in commercial stay-on products was 8, and 6 of these were considered to be elicited by Compositae plants.

Even though the majority of participants had multiple contact allergies, positive patch test reactions were not restricted to this group. 3 of 12 chamomile-sensitive subjects had no other contact

allergy (at the beginning of the study; patient no. 8 developed a serious occupationally related flare of her hand eczema 2 years later and tested positive to 3 standard series allergens in 2005) and they constituted the few chamomile tea-positive, even though another 5 persons had doubtful positive reactions to chamomile tea. Hausen reported that chamomile tea contained SLs (10), and this was apparently supported by a recent Swedish study (11): among 19 Compositae-allergic persons, 11 tested positive to a stock solution of German chamomile tea (11). This prevalence is considerably higher than the 1 detected in our selected chamomile-sensitive patients, which may be because of different patch test concentrations. We tested with tea in an ordinary drinking concentration and furthermore, the patients were tested on 11 separate days with freshly made tea, allowing for varying concentrations of possible allergens. In contrast to this, the Swedish patients were tested with an extract made from 1.5 g dried herbs in 50 ml of boiling water, which, after evaporation of the water and reconstitution with 1 ml of water, was called the stock solution (11). The strongly positive reactions to this stock solution in the Swedish and the 5 doubtful positive reactions in our study support this explanation.

The Roman chamomile extract elicited fewer positive reactions. This is quite intelligible in view of the fact that Roman chamomile is not native to and hence, less used as a medicinal plant in Denmark compared with the UK, France, and Belgium (12). Nevertheless, 3 patients tested positive to Roman chamomile tea. The results emphasize, like previous reports on contact allergy from German or Roman chamomile teas, that the application of teas to the skin for medical purposes may sensitize or elicit dermatitis in presensitized individuals (13, 14).

The fact that weak herbal teas may elicit allergic contact dermatitis both clinically by ingestion or topical use and experimentally by patch testing suggests that allergens other than SLs may be involved as the latter are often insoluble in water (11–13). Thus, the SLs helenalin and carabron from *A. montana* are slightly soluble or insoluble in water, respectively, but soluble in alcohol (12). The chemical analyses are in accordance with this as only trace amounts of SLs were detected in the aqueous solutions of arnica, whereas SLs appeared to be absent in the infusions of German chamomile and marigold. Nevertheless, patient no. 15, who was an atopic individual with several contact and type I allergies, tested positive to the arnica infusion with trace amounts of SLs and the SL-free infusion of marigold (and doubtful positive to the chamomile infusion) as well as positive

to Compositae mix, but negative to SL mix (Table 9). Even though this pattern of reaction to the mixes is a common finding in Compositae-sensitive persons, it may also reflect sensitization to allergens of a non-sesquiterpenoid structure present in the crude flower extract. Patient no. 7, who was tested with the same ingredients, had doubtful positive reactions to the arnica and chamomile extracts, indicating the presence of weak allergens or a low concentration of allergens.

Alternatively, patient no. 15, who had positive skin prick and histamine release tests to iceberg lettuce and sunflower seeds, tested positive to water-soluble allergens usually involved in immediate type hypersensitivity. Finally, the patient may have tested positive to the traces of SLs in the arnica infusion. The allergens of marigold are unknown (12), but an Austrian study showed a high prevalence of patch test positivity to an ether extract of marigold, most often of current relevance (15).

The mixture of tinctures, on the other hand, elicited positive reactions in 2 of 3 patients tested, and the third was eventually considered to have been included erroneously because of angry back reactions in the primary testing (Tables 8 and 9). The mixture contained, among other compounds, tinctures of 4 Compositae plants, but only the arnica tincture comprised minute amounts of several SLs. Both persons (patients nos. 9 and 18) had strongly positive (2+) reactions to the mixture, and when testing with the individual tinctures, except those containing German chamomile and arnica, patient no. 9 had a 2+ reaction to *Hamamelis* tincture, while patient no. 18 tested negative/doubtful positive to all of them. As both subjects were strongly arnica-sensitive, the arnica tincture may be considered a possible (patient no. 9) and a probable (patient no. 18) contributing factor to the mixed tincture reactions.

Likewise, the CO₂-extract of arnica and sunflower contained several SLs, which were mainly helenalin, dihydrohelenalin and short chain esters of dihydrohelenalin of *A. montana*. In accordance with this 2 of 3 arnica-sensitive subjects tested positive to the extract; the third was a doubtful positive (Table 9).

Blue chamomile flower oil was positive in 1 person only, and she had tested positive to peppermint and spearmint mixes as well as lemon and sandalwood oil previously. Thus, the chamomile oil reaction probably represented cross- or concomitant reactivity between essential oils rather than sensitization to specific Compositae allergens, which is supported by the fact that no SLs were detected in the oil. Alternatively, the patient

may be sensitized to other allergenic compounds in the oil such as polyacetylenes or bisabolol derivatives (12, 16).

H. virginiana (witch hazel) is a shrub or small tree from the Hamamelidaceae family of plants. Witch hazel extract is attributed astringent, antiseptic, anti-inflammatory, local styptic, and vasoconstrictive properties (17–19). It has been used topically for inflammation of the skin and mucous membranes, and today it appears in cosmetic products as well as oral medicine for chronic venous insufficiency (17, 20). Contact dermatitis from the extract has been reported a few times since the first publication in 1937, most recently and convincingly by Granlund who described a case of periorbital dermatitis from witch hazel distillate used in an eye gel (21, 22). Bruynzeel et al., on the other hand, detected 4 cases of positive patch test reactions to an ointment containing *Hamamelis* extract among 1032 patients tested; 2 of the 4 patients tested positive to 2 or 3 Compositae-containing ointments as well, but this was ascribed to their reaction to 1 of the constituents of the ointment bases (23). The witch hazel reaction in our patient was of unknown relevance, and likewise the possibility of cross-reactivity between witch hazel and Compositae extracts is unknown. Unfortunately, the patients in the study of Bruynzeel et al. were not patch tested with plant extracts, which may have been a contributing factor to the positive ointment reactions (23).

Avocado oil, obtained from the avocado pear (*Persea gratissima* = *Persea Americana* Mill.) of the Lauraceae family of plants, is an emollient and conditioning agent used in skin and hair care products. The oil seems to be a rare sensitizer: de Groot et al. detected 1 case of sensitization to avocado oil used in a sunscreen among 119 persons with cosmetic-related contact dermatitis (24, 25), and Barbaud et al. reported a case of positive patch test reaction to avocado oil in a patient who had taken avocado oil-containing capsules for years without any adverse effects (26). It is, therefore, surprising to find 2 cases of possibly relevant sensitization in this small study. Avocado oil is obtained from the dehydrated, sliced flesh of the avocado fruit, which also contains numerous terpenes (24, 27). Both patients (nos. 7 and 15) tested positive to fragrance terpenes, and cross-reactivity between these and minute amounts of terpenes in the avocado oil is 1 possible explanation (27). Some members of the laurel family contain SLs, but apparently these have not been detected in avocado fruit or flowers (27, 28). Incidentally, patient no. 15 had a positive prick test to latex and avocado.

In spite of the limitations of the study, especially concerning the selected testing of only some of the potential allergenic compounds from some products, the conclusion is that Compositae-sensitive patients should be warned against the topical use of Compositae-containing cosmetics and herbal remedies, including herbal teas and other aqueous extracts and essential oils. The Compositae-monosensitized may react to plant extracts, and the patients with multiple contact allergies may react to preservatives, emulsifiers, fragrance chemicals and plant extracts. Even though the plant-derived compounds comprised the largest group of allergens in the present study, many Compositae-sensitive persons have other contact allergies, and therefore the warning is appropriate although not (always) for the entirely right reason: fragrances, emulsifiers, and preservatives may pose just as great a risk as the plant allergens.

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References

- Paulsen E. Contact sensitization from Compositae-containing herbal remedies and cosmetics. *Contact Dermatitis* 2002; 47: 189–198.
- Wahlberg J E, Lindberg M. Patch testing. In: *Contact Dermatitis*, 4th edition, Frosch P J, Menné T, Lepoittevin J-P (eds): Berlin-Heidelberg, Springer-Verlag, 2006: 365–390.
- Kos O, Lindenmeyer M T, Tubaro A, Sosa S, Merfort I. New sesquiterpene lactones from Arnica tincture prepared from fresh flowerheads of *Arnica montana*. *Planta Med* 2005; 71: 1044–1052.
- Bilia A R, Bergonzi M C, Mazzi G, Vincieri F F. Development and stability of semisolid preparations based on a supercritical CO₂ Arnica extract. *J Pharm Biomed Anal* 2006; 41: 449–454.
- Tsai L, Highet R J, Herz W. The mass spectra of pseudoguaianolides related to helenalin. *J Org Chem* 1969; 34: 945–948.
- Poplawski J, Holub M, Samek Z, Herout V. Arnicolides – sesquiterpenic lactones from the leaves of *Arnica montana* L. *Collect Czech Chem Commun* 1971; 36: 2189–2199.
- Herrmann H-D, Willuhn G, Hausen B M. Helenalinmethacrylate, a new pseudoguaianolide from the flowers of *Arnica montana* L. and the sensitizing capacity of their sesquiterpene lactones. *Planta Med* 1978; 34: 299–304.
- Repčák M, Imrich J, Garčár J. Quantitative evaluation of the main sesquiterpenes and polyacetylenes of *Chamomilla recutita* essential oil by high-performance liquid chromatography. *Phytochem Anal* 1999; 10: 335–338.
- Lomholt H, Rastogi S C, Andersen K E. Allergic contact dermatitis from sodium dihydroxycetyl phosphate, a new cosmetic allergen? *Contact Dermatitis* 2001; 45: 143–145.
- Hausen B M. Lorbeer-Allergie. *Dtsch Med Wochenschr* 1985; 110: 634–638.
- Lundh K, Hindsén M, Gruvberger B, Möller H, Svensson Aa, Bruze M. Contact allergy to herbal teas derived from Asteraceae plants. *Contact Dermatitis* 2006; 54: 196–201.
- Hausen B M, Vieluf I K. *Allergiepflanzen/Pflanzenallergene. Handbuch und Atlas der allergieinduzierenden Wild- und Kulturpflanzen*, 2nd edition. Landsberg/München, Ecomed Verlagsgesellschaft mbH, 1997.
- Pereira F, Santos R, Pereira A. Contact dermatitis from chamomile tea. *Contact Dermatitis* 1997; 36: 307.
- Rodríguez-Serna M, Sánchez-Motilla J M, Ramón R, Aliaga A. Allergic and systemic contact dermatitis from *Matricaria chamomilla* tea. *Contact Dermatitis* 1998; 39: 192–193.
- Reider N, Komericki P, Hausen B M, Fritsch P, Aberer W. The seamy side of natural medicines: contact sensitization to arnica (*Arnica montana* L.) and marigold (*Calendula officinalis* L.). *Contact Dermatitis* 2001; 45: 269–272.
- Wilkinson S M, Hausen B M, Beck M H. Allergic contact dermatitis from plant extracts in a cosmetic. *Contact Dermatitis* 1995; 33: 58–59.
- Bedi M K, Shenefelt P D. Herbal therapy in dermatology. *Arch Dermatol* 2002; 138: 232–242.
- Gloor M, Reichling J, Wasik B, Holzgang H E. Antiseptic effect of a topical dermatological formulation that contains *Hamamelis* distillate and urea. *Forsch Komplementarmed Klass Naturheilkd* 2002; 9: 153–159.
- Hughes-Formella B J, Bohnsack K, Rippke F, Benner G, Rudolph M, Tausch I, Gassmueller J. Anti-inflammatory effect of *Hamamelis* lotion in a UVB erythema test. *Dermatology* 1998; 196: 316–322.
- Kiken D A, Cohen D E. Contact dermatitis to botanical extracts. *Am J Contact Dermat* 2002; 13: 148–152.
- Michthell J C, Rook A. *Botanical Dermatology*. Vancouver, Greengrass, 1979.
- Granlund H. Contact allergy to witch hazel. *Contact Dermatitis* 1994; 31: 195.
- Bruynzeel D P, van Ketel W G, Young E, van Joost T, Smeenk G. Contact sensitization by alternative topical medicaments containing plant extracts. *Contact Dermatitis* 1992; 27: 278–279.
- de Groot A C, van der Meeren H L M, Weyland J W. Contact allergy to avocado oil in a sunscreen. *Contact Dermatitis* 1987; 16: 108–109.
- de Groot A C, Bruynzeel D P, Bos J D, van der Meeren H L M, van Joost T, Jagtman B A, Weyland J W. The allergens in cosmetics. *Arch Dermatol* 1988; 124: 1525–1529.
- Barbaud A, Trechot P, Reichert-Penetrat S, Commun N, Schmutz J L. Relevance of skin tests with drugs in investigating cutaneous adverse drug reactions. *Contact Dermatitis* 2001; 45: 265–268.
- Pino J A, Marbot R, Rosado A, Fuentes V. Volatile components of avocado (*Persea americana* Mill.) cv. Moro grown in Cuba. *J Essent Oil Res* 2004; 16: 139–140.
- Kruthiventi A K, Krishnaswamy N R. Constituents of the flowers of *Persea gratissima*. *Fitoterapia* 2000; 71: 94–96.

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