

Pyrethrins and Pyrethrosin Content in Commercial Allergen Extracts

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Background: Pyrethrins are the insecticidally active components of pyrethrum extract, derived from flowers of *Chrysanthemum cinerariifolium* and used in commercial and consumer insecticide products. Most dermal testing performed with pyrethrum extracts was done before current refined pyrethrum concentrate became available (before 1967).

Objective: We analyzed presently commercially available pyrethrum allergen extracts to determine the concentration of pyrethrins and the putative sensitizer pyrethrosin.

Methods: Six commercial pyrethrum allergen extracts were purchased from four major allergen suppliers and analyzed for pyrethrin I and pyrethrosin by using a capillary gas chromatograph equipped with a flame ionization detector.

Results: The commercial pyrethrum allergen extracts contained no detectable pyrethrins or pyrethrosin. In comparison, the pyrethrum standard provided by the McLaughlin Gormely King Company, a major refiner of pyrethrum, contained 20% pyrethrins and 0.49% pyrethrosin. No compounds observed in the chromatogram of the refined pyrethrum concentrate were present in the allergen extracts.

Conclusions: Caution should be used when interpreting the results of tests performed with current pyrethrum allergen extracts because pyrethrins and pyrethrosin may not be present. Moreover, unknown components such as high-molecular-weight proteins or other impurities that may cause dermal reactions could be present in significant amounts.

PYRETHRINS are the insecticidally active components of pyrethrum extract, derived from the flowers of *Chrysanthemum cinerariifolium*, which is grown as a crop mainly in East Africa (Kenya, Tanzania, and Rwanda) and Tasmania, Australia, and used in commercial and consumer insecticide products. The literature includes numerous reports of dermatitis from pyrethrum and pyrethrins, but comprehensive evidenced-based reviews fail to document a clear causal association between pyrethrins and either contact urticaria or allergic contact dermatitis.^{1,2} Much of the dermal testing performed with pyrethrum extracts and reported in the literature was done before current refined pyrethrum concentrate was available (before 1967).² It is likely that some of the subjects in older studies were patch-tested either with irritating levels of

pyrethrins or with samples containing unknown impurities, leading to false-positive reactions.

Determining the appropriateness of an allergen sample requires an understanding of the commercial growing, harvesting, and refining of pyrethrins.² African pyrethrum harvesting is done by hand and reflects the asynchronous flowering pattern of the African plants—buds and immature flowers are present along with mature blooms. In contrast, the Australian flowering pattern is synchronous; all flowers mature at the same time and can thus be harvested mechanically. The flowers are then field dried.

The dried flowers are coarsely ground, and the grist is extracted by percolation with low boiling isoalkanes such as hexanes. The resulting solution is filtered to leave a viscous dark greenish brown liquid oleoresin.

The oleoresin is partitioned by liquid partitioning into a polar low-boiling agent such as methanol (East African technique) or subcritical liquid carbon dioxide (Australian technique).² The aliquot containing diluted pyrethrins is decolorized with active carbon and filtered. The partitioning agent is then removed by distillation.

The refining stage substantially removes triglycerides, high-molecular-weight plant waxes, plant pigments, and polymers. Commercial refining currently takes place in East Africa, Australia, and North America; although the techniques vary somewhat, a similar product is produced

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from each location. These processes evolved throughout the 1950s and 1960s. By 1967, all of the refiners were using processes that resulted in a highly refined extract fundamentally different from that produced in the first half of the century.

The residue containing highly purified pyrethrins is adjusted to a desired approved concentration by diluting it with a hydrocarbon diluent such as a commercially available isoalkane. It is then suitable for formulating (eg, as a water-emulsifiable concentrate) by adding inert emulsifiers and wetting agents to produce a commercial product.

Several commercial allergen suppliers offer pyrethrum preparations for use in diagnostic testing. Descriptions of the procedures by which our samples were made were not readily available from the suppliers, so it is uncertain whether the samples are representative of the pyrethrins actually encountered by the public. Preparations are offered in glycerin, petrolatum, and saline solution.

Objective

We analyzed presently commercially available pyrethrum allergen extracts to determine the concentration of pyrethrins³ and pyrethrosin (the putative sensitizer speculated to elicit type IV reactions).

Methods

Six commercial pyrethrum allergen extracts were purchased from four major allergen suppliers. Samples were analyzed in duplicate; 4 mg of dibutyl phthalate were added to 0.2 g of each sample, which were then diluted to volume with methanol. The analysis was performed with a capillary gas chromatograph (Hewlett Packard 6890,

Agilent Technologies, Inc., Santa Clara, CA) equipped with a flame ionization detector (300°C) and a cool on-column injection port. The column was an HP-1 25 m × 0.32 mm capillary column with a 0.52 µm film thickness (programmed from 100°–280°C at 5°C/min). Quantitation was by comparison with peak height ratios, with a refined pyrethrum concentrate (supplied by McLaughlin Gormley King Company [MGK], Golden Valley, MN)⁴ serving as the internal standard.

Results

Our analysis showed that the commercial pyrethrum allergen extracts contained no detectable pyrethrins or pyrethrosin (Table 1). In comparison, the MGK standard contained 20% pyrethrins and 0.49% pyrethrosin. No compounds seen in the chromatogram of the refined pyrethrum concentrate were present in the allergen extracts. The following several factors could account for this:

- The technique used by the allergen sample suppliers may not extract significant amounts of pyrethrins or pyrethrosin from the raw flowers; such would be the case if simple aqueous extractions were performed (given the low water solubility of pyrethrins and pyrethrosin).
- The pyrethrum flowers used had low levels of pyrethrins because the concentrations are dependent on climate, maturity, time of harvest, etc.³
- Because they are susceptible to hydrolysis, photolysis, and degradation due to heat,⁴ pyrethrins could have been extracted but destroyed as a result of handling or storage techniques.

The normal commercial extraction and refining process and the allergen extract processes seem to isolate mutually exclusive sets of compounds.

Table 1. Determination of Pyrethrins and Pyrethrosin Content in Commercially Available Allergen Samples*

Supplier [†]	Sample [‡]	Vehicle	Purity (w/v)	Quantity	Intended Use	Pyrethrins	Pyrethrosin
1	A	50% glycerin	1:20	10 mL	P	< LOD	< LOD
2	A	Aqueous	1:10	10 mL	ID	< LOD	< LOD
	B	50% glycerin	1:20	10 mL	P	< LOD	< LOD
3	A	Aqueous	1:10	10 mL	ID	< LOD	< LOD
	B	50% glycerin	1:20	10 mL	P	< LOD	< LOD
4	A	Petrolatum	1	5 g	P	< LOD	< LOD
MGK	Standard	None	NA	NA	NA	20%	0.49%

ID = intradermal; LOD = limit of detection; MGK = McLaughlin Gormley King Company; NA= not applicable; P = patch; w/v = weight per volume.

*Limits of detection: pyrethrins I = 250 ppm; pyrethrins II = 1,000 ppm; pyrethrosin = 25 ppm.

‡All analyses were conducted prior to the expiration date of the sample.

Conclusions

Caution should be used when interpreting the results of tests performed with current pyrethrum allergen extracts from some allergen suppliers. Unexpectedly, pyrethrins and pyrethrosin may not be present. Moreover, unknown components such as high-molecular-weight proteins or other impurities that may cause dermal reactions could be present in significant amounts in aqueous extracts of pyrethrum as prepared by some allergen suppliers. It has been demonstrated that aqueous pyrethrum flower extracts contain significant amounts of nitrogen in the form of high-molecular-weight nondialyzable molecules.⁵ Moreover, these extracts have been shown to elicit strong reactions in guinea pigs previously sensitized to suspended ground pyrethrum flowers. In contrast, commercially refined pyrethrum extracts provoked no response.⁵

These findings stress the need for the development of a dermal diagnostic pyrethrum standard that reflects current processing methods for insecticide products (as opposed to allergen samples) and the determination of a non-irritating dose level for the diagnostic standard for both patch testing and immediate-type testing.

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