COMPARISON AND CHARACTERIZATION OF GARLIC 
(Allium sativum L.) BULBS EXTRACTS AND THEIR EFFECT ON 
MORTALITY AND REPELLENCY OF Tetranychus urticae KOCH 
(ACARI: TETRANYCHIDAE)

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INTRODUCTION

Tetranychus urticae Koch is a pest that causes great economic losses because it attacks a wide range of host plants. Also, some populations have developed resistance against commercial acaricides. This work compares different extracts from garlic (Allium sativum L.) bulbs measuring their toxicity and repellency effects on T. urticae. Extracts were obtained using as solvents CO2 in supercritical conditions (CSC), ethanol and petroleum ether using Soxhlet and soaking methods and soaking in water. The supercritical fluid extraction technique allows for obtaining extracts at low temperature using high pressures, avoiding compound degradation and making possible solvent separation by exposing the extract at room temperature. Mites were bred on bean plants (Phaseolus vulgaris L.) in a laboratory environment in Laureles Campus, Universidad Pontificia Bolivariana, Medellín, Colombia. Mortality was evaluated putting 10 mites in P. vulgaris 3 cm diameter leaf discs previously submerged in the extract. The lowest mean lethal concentrations (LC50) were obtained with the CSC method (8118.8, 5410.5, 2820.6 mg kg−1 at 24, 48 and 72 h, respectively). Extracts were characterized using gas chromatography/mass spectrometry (GC/MS), finding vinyl dithiins (63.11%), diallyl disulfide (10.88%) and diallyl trisulfide (10.4%) as the main compounds in the CSC extract. The main conclusion is that, in comparison with other techniques of extraction from garlic bulbs used in this study, supercritical fluids allow for obtaining extracts with a higher concentration of biologically active compounds against T. urticae.

Key words: two-spotted spider mite, plant extracts, miticidal effect, Liliaceae.
Some plants with acaricide and/or repellent effect against the mite are: *A. sativum* in an aqueous solution (Boyd and Alverson, 2000), *Artemisia absinthium* L. and *Tanacetum vulgare* L. (Chiasson et al., 2001), *Piper longum* L. (Park et al., 2002), *Trichilia pallida* Swartz and *Melia azedarach* L. (Castiglioni et al., 2002), *Azadirachta indica* A. Juss. (Castiglioni et al., 2002; Knapp and Kashenge, 2003; Chiasson et al., 2004); *Satureja hortensis* L., *Ocimum basilicum* L. and *Thymus vulgaris* L. (Aslan et al., 2004); *Carum carvi* L., *Cymbopogon nardus* (L.) Rendle, *Eucalyptus citriodora* Hook, *Mentha pulegium* L., *Mentha piperita* L., *Mentha spicata* L. (Choi et al., 2004), *Chenopodium ambrosioides* var. ambrosioides (Chiasson et al., 2004). Some organisms on which the insecticide or repellent action of garlic have been tested are: *Acanthoscelides obtectus* Say (Regnault-Roger and Hamraoui, 1993); *Tribolium castaneum* and *Sitophilus zeamais* (Ho et al., 1996; Chiam et al., 1999; Huang et al., 2000); *Cydia pomonella* L. (Landolt et al., 1999); *Lipaphis erysimi* Kaltenbach and *Dysdercus cingulatus* (Bandyopadhyay et al., 2001; Banerjee et al., 2004); *Meloidogyne incognita* (Chitwood, 2002); *Reticulitermes santonensis* and *Reticulitermes grassei* (Auger et al., 2004); and *Attagenus unicolor japonicus* (Han et al., 2006).

The traditional extraction methods can cause interactions between solvents and extracts, producing modifications in the original structure of the compounds. Besides, there is the risk of chemical residues that affects the ecological behavior of the extract. Extraction with supercritical fluids is a highly beneficial alternative to remove these problems. A supercritical fluid is any substance in conditions of pressure and temperature higher than its critical point; it is a quasi-state with intermediate properties between a liquid and a gas. Supercritical fluids is a highly beneficial alternative to remove these problems. A supercritical fluid is any substance in conditions of pressure and temperature higher than its critical point; it is a quasi-state with intermediate properties between a liquid and a gas. Supercritical CO$_2$ presents the best extraction conditions, with less interaction with the extract and low environmental impact (McClain, 2000).

The objective of this study was to compare the effects of different extract obtention methods from bulbs of *A. sativum* (supercritical CO$_2$, water, ethanol and petroleum ether as solvents) on *T. urticae*. Each extract was characterized through CG/MS to identify the compounds and determine which have possibilities of biological action on mites.

**MATERIALS AND METHODS**

The study was done in 2006 at the Universidad Pontificia Bolivariana (Campus Laureles), Medellín (6°14’ N, 75°35’ W), Antioquia, Colombia, at an average altitude of 1450 m.a.s.l. and an average temperature of 22 ± 1 °C (IDEAM, 1999). The initial colony of mites was supplied by the Universidad Militar Nueva Granada, Cajicá Campus, Cundinamarca, Colombia. To maintain the colony and carry out the tests, bean plants cv. Cargamanto, were seeded under laboratory conditions, so that germination and growth occurred in isolation. A tray was seeded weekly with approximately 12 bean seeds. When the plants were 4 weeks old, they were inoculated with leaves infested with mites. The plants were replaced every 4 weeks. The plants were irrigated daily and organic fertilizer was applied at seeding. Pest control was done manually in order to not use synthetic chemical products and thus avoid introducing error factors to the experiment.

Garlic bulbs from a cultivar known as “China white” were obtained in the local market, an option that has been taken by other researchers (Kuo and Ho, 1992; Calvey et al., 1994; Abu-Lafi et al., 2004; Kimbaris et al., 2006; Han et al., 2006). Subsequently, the bulbs were selected, cleaned, cut and mashed manually in the laboratory. The product obtained was used for the extractions. For the extraction with supercritical CO$_2$ gas (UN 2817, Agafano, Medellín, Colombia) was used as the solvent under supercritical conditions in an extractor (SFE1000, Thar Technologies, Pittsburgh, USA) located in the private company Colflavor S.A., Envigado, Antioquia. The temperature in the extractor was 40 °C and the pressure and CO$_2$ flow were 25.33 MPa and 30 g min$^{-1}$, respectively (Ryback et al., 2004). The extraction time was 1.5 h. For extraction using the Soxhlet method, 100 g of sample was used, and petroleum ether 35-60 and ethanol at 99.7% were used as solvents, for 8 h. For soaking at room temperature, 100 g of sample in a hermetically sealed container was submitted to agitation for 24 h, using the same solvents as used with Soxhlet. Finally, the extractions were filtered and the solvents separated at low temperature in a rotary evaporator (Waterbath-B-480, Büchi Labortechnik AG, Flawil, Switzerland). Soaking was also carried out in water. Unlike the previously described methods, in the latter case, the concentration was determined making a mass balance. To prepare each treatment, the solvents were removed exhaustively to avoid their incidence in the tests. The extracts prepared were: supercritical CO$_2$ (CSC), soaking with petroleum ether (MEP), Soxhlet with ethanol (SAE), Soxhlet with petroleum ether (SEP), soaking with ethanol (MAE), and soaking with water (MA). Before use the extracts were stored in a refrigerator in hermetically sealed amber-colored
To determine mortality with CSC extracts, concentrations of 2,500, 5,000 and 10,000 mg kg\(^{-1}\) were prepared; as well, concentrations of 10,000, 25,000, 50,000, 100,000, 150,000 and 200,000 mg kg\(^{-1}\) were prepared for the MEP, MAE, SEP and SAE extracts. Concentrations of 68,500, 45,700, 22,800, 11,400 and 4,600 mg kg\(^{-1}\) were prepared for the MA extract. The extract was solved in distilled water. Owing to conditions inherent to the soaking process and the concentration of the solvent, extracts of over 76,800 mg kg\(^{-1}\) were not obtained. Thus 34 treatments were obtained, with three replicates of each one. Concentrations of 1,000, 2,000 and 5,000 mg kg\(^{-1}\) were used with all the extracts for repellence tests. A completely random design was used, with 24 treatments and three replicates of each, 18 treatments (six extracts with three concentrations), the absolute control (distilled water) and the positive control, abamectina 75 mg kg\(^{-1}\) (CATOMBE 1.8 EC, Químicos Oma, Colombia). The concentrations were determined after preliminary tests to establish the ranges of action on the mites.

For the mortality tests eight 3-cm diameter disks of young leaves of bean were immersed in each treatment, and then deposited on Petri plates with filter paper moistened with distilled water. After drying, 10 adult mites were placed in each disk and were covered. Mortality was evaluated at 24, 48 and 72 h and a mite was considered dead when it presented no movement after being poked by a brush bristle (Chiaison et al., 2001; 2004; Castiglioni et al., 2002; Kwon and Ahn, 2002a; 2002b; Kim et al., 2003; 2004a; 2004b; Choi et al., 2004). The percentage of mortality was calculated considering the data obtained from the absolute control and the treatments corrected with Abbott’s formula (1925).

For the repellence bio-tests the same procedure was used as for the mortality tests, except that over half of each 3 cm disk a 14 mm leaf disk without treatment was placed, and 10 adult mites were placed on this smaller disk. The mites that remained there were counted at 2, 3 and 4 h (Boyd and Alverson, 2000). The information gathered in the repellence tests was analyzed with repeated measurement using the PROC MIX module from the SAS program (SAS Institute, 1999), with which the variance analysis was carried out. Significant differences were identified with the Minimal Significant Differences test. The mean lethal concentration (LC\(_{30}\)) was determined using the PROBIT module of the SAS program, with the probit method proposed by Bliss (1934) and developed by Finney (1947).

The extracts were re-dissolved into their respective solvents for the chromatographic analysis. The CSC extract was dissolved in dichloromethane (Calvey et al., 1994). For the MA extract, an equal quantity of dichloromethane was added in order to extract the sulphurated compounds of low to medium polarity present in this extract (Kuo and Ho, 1992). The solvents were eliminated before using the mass detector. The characterization was done by CG/MS (6890N Network GC System, Mass selective detector 5973 Network and Injector 7683 Series; Agilent Technologies, Palo Alto, California, USA) in the laboratory of instrumental analysis of the Universidad Nacional de Colombia, Medellín Campus. An HP 5 fused silica capillary column was used (30 m x 0.25 mm, film thickness of 0.25 μm) (Abu-Lafi et al., 2004; Kimbaris et al., 2006). For detection it was used ionization energy of 70 eV. The temperatures of the injector and the detector were fixed at 220 and 290 °C, respectively. An helium flow used as a gas drag was 1 mL min\(^{-1}\) (Kimbaris et al., 2006). The oven was programmed to begin at 40 °C for 2 min, increasing 2 °C every 2 min (Block, 1992) until 300 °C, which was maintained for 20 min (Abu-Lafi et al., 2004). The compounds were identified with the NIST 98 library incorporated into the equipment.

**RESULTS AND DISCUSSION**

**Mortality**

The CSC extract was the most toxic, followed by MEP (Table 1). Mortalities of less than 40% were obtained with MA in the majority of the cases, because of which it registered LC\(_{30}\); it should be noted that the concentration of the extract could not be increased because it is not obtained in pure form but diluted in water, at a concentration that does not exceed on average 76,800 mg kg\(^{-1}\). The extracts that showed greater mortality were obtained under conditions of low temperatures, which suggests that these conditions favor obtaining compounds with greater acaricide effect than those that can be obtained at higher temperatures. The solvent polarity also appears to influence the results, given that the CO\(_2\) and petroleum ether are non-polar solvents. These two extracts have similar percentages of diallyl sulfide (CSC: 23.058% and MEP: 24.842%) and disulfide (CSC: 12.537% and MEP: 14.798%), but present major differences in vinyl dithiins (CSC: 63.11% and MEP: 10.173%). With regard to the disulfides, the two containers and injected with highly purified nitrogen gas (Agafano, Medellín, Colombia).

The information gathered in the repellence bio-tests was analyzed with repeated measurement using the PROC MIX module from the SAS program (SAS Institute, 1999), with which the variance analysis was carried out. Significant differences were identified with the Minimal Significant Differences test. The mean lethal concentration (LC\(_{30}\)) was determined using the PROBIT module of the SAS program, with the probit method proposed by Bliss (1934) and developed by Finney (1947).
The compounds found in greater quantity in the MEP extract are methyl 2-propanol disulfide, dimethyl trisulfide, diallyl disulfide, 2-ethylidene[1,3]dithiane, and di-2-propenyl trisulfide and 3-vinyl-1,2-dithiociclohex-5-ene (Table 3). These could be responsible for the repellent effect owing to their volatility and strong odor. The MA extract has a low percentage of such compounds and the CSC has only 1.657% methyl 2-propanol disulfide and trisulfide, but an appreciable concentration of diallyl disulfide (10.88%). These two extracts were the least repellent. These results are better than those obtained by Boyd and Alverson (2000), who used concentrations of 100, 50, 20 and 10% of Garlic Barrier AG (a commercial insect repellent described as aqueous garlic [Garlic Barrier, 1999]), an aqueous garlic juice produced in the laboratory and a mixture of mineral oil and garlic powder. Significant repellence was observed with the commercial product at 100% in all the times, and for 30 and 45 min at 50%. This suggests that this type of extract should be used in almost pure form in order to have any effect.

From the statistical point of view, the use of any extract as a repellent of *T. urticae* in the concentrations used in this study is the same (with the exception of the aqueous and the CSC extracts at 1000 and 2000 mg kg\(^{-1}\)). As well, it was found that there is a statistically significant relationship between the percentage of acyclic sulphurated compounds and maximum repellence obtained at 4 h (\(r^2 = 87.2046\) and \(p = 0.0064\)). This correlation suggests that acyclic compounds present in the extracts are, in great part, responsible for the repellence of mites. To define a possible commercial product or to carry out further tests it will be necessary to analyze factors such as the performance of the extraction, solvent cost, saving of energy and processing time, among others.

### Compounds identified and their influence on mortality and repellence

A number of important sulphurated compounds were identified in the majority of the extracts between 10 and 63 min (retention time) (Figures 1 and 2). After
Table 2. Repellence percentages on *Tetranychus urticae* of each garlic bulb extract in each evaluated time.

<table>
<thead>
<tr>
<th>Extraction method</th>
<th>Concentration (mg kg(^{-1}))</th>
<th>0.25</th>
<th>0.5</th>
<th>0.75</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
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<td><strong>CSC</strong></td>
<td>1000</td>
<td>36.67a</td>
<td>30.00a</td>
<td>26.67a</td>
<td>26.67a</td>
<td>30.00a</td>
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<td>16.67a</td>
</tr>
<tr>
<td></td>
<td>2000</td>
<td>36.67a</td>
<td>33.33a</td>
<td>36.67a</td>
<td>33.33a</td>
<td>30.00a</td>
<td>26.67a</td>
<td>26.67ab</td>
</tr>
<tr>
<td></td>
<td>5000</td>
<td>83.33c</td>
<td>70.00cde</td>
<td>63.33bc</td>
<td>66.67bcdef</td>
<td>66.67bcdef</td>
<td>66.67bcdef</td>
<td>63.33def</td>
</tr>
<tr>
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<td>90.00cde</td>
<td>90.00cde</td>
<td>90.00cde</td>
<td>90.00cde</td>
<td>90.00cde</td>
<td>90.00cde</td>
</tr>
<tr>
<td></td>
<td>2000</td>
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<td>100.00cde</td>
<td>100.00cde</td>
<td>100.00cde</td>
<td>93.30cde</td>
<td>93.30cde</td>
<td>96.70h</td>
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<tr>
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<td>86.70cde</td>
<td>86.70cde</td>
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<tr>
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<td>93.30cde</td>
<td>96.70cde</td>
<td>93.30cde</td>
<td>93.30cde</td>
<td>93.30cde</td>
<td>90.00fgh</td>
</tr>
<tr>
<td></td>
<td>2000</td>
<td>93.30c</td>
<td>86.70cde</td>
<td>90.00cde</td>
<td>90.00cde</td>
<td>76.70cde</td>
<td>66.70bcdef</td>
<td>70.00defg</td>
</tr>
<tr>
<td></td>
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<td>86.70c</td>
<td>76.70cde</td>
<td>86.70cde</td>
<td>86.70cde</td>
<td>83.30cdef</td>
<td>83.30cdef</td>
<td>83.30cdef</td>
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<tr>
<td><strong>SEP</strong></td>
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<td>76.70bcdef</td>
<td>76.70bcdef</td>
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<tr>
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<td>93.30cde</td>
<td>93.30cde</td>
<td>83.30cde</td>
<td>83.30cde</td>
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<td>86.70cdef</td>
<td>73.30cdef</td>
<td>70.00defg</td>
<td>73.30defgh</td>
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<tr>
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<td>80.00cde</td>
<td>76.70bcdef</td>
<td>76.70bcdef</td>
<td>70.00cde</td>
<td>70.00cde</td>
<td>63.30defgh</td>
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<tr>
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<td>40.00ab</td>
<td>40.00ab</td>
<td>40.00ab</td>
<td>33.30abc</td>
</tr>
<tr>
<td></td>
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<td>53.30ab</td>
<td>53.30abc</td>
<td>53.30ab</td>
<td>53.30abc</td>
<td>50.00abc</td>
<td>50.00abc</td>
<td>50.00bcd</td>
</tr>
<tr>
<td></td>
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<td>76.70bc</td>
<td>70.00cde</td>
<td>70.00cde</td>
<td>63.30abcde</td>
<td>60.00cde</td>
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<tr>
<td>Absolute control</td>
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<td>66.70bcd</td>
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<td>53.30bcd</td>
<td>46.70abc</td>
<td>33.30ab</td>
<td>23.30ab</td>
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<tr>
<td>Positive control (abamectin)</td>
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<td>73.33cde</td>
<td>70.00bcd</td>
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<td>66.67bcddef</td>
<td>66.67cdef</td>
<td>66.67defg</td>
</tr>
</tbody>
</table>

Data followed by the same letter does not have a significant difference according to the minimum significant difference test (P < 0.05).

CSC: supercritical CO\(_2\); MEP: soaking with petroleum ether; SAE: Soxhlet with ethanol; SEP: Soxhlet with petroleum ether; MAE: soaking with ethanol; MA: soaking with water.

![Figure 1. Chromatogram with peaks that indicate identified compounds from supercritical CO\(_2\) extract.](image-url)
### Table 3. Comparison between most found compounds in each extract.

<table>
<thead>
<tr>
<th>Name of the compound</th>
<th>Peak area with each extraction method</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CSC</td>
</tr>
<tr>
<td>Methyl 2-propanol disulfide</td>
<td>1.66</td>
</tr>
<tr>
<td>Dimethyl trisulfide</td>
<td>Nd</td>
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<tr>
<td>Diallyl disulfide</td>
<td>10.88</td>
</tr>
<tr>
<td>2-Ethylidene[1,3]ditianate</td>
<td>Nd</td>
</tr>
<tr>
<td>Methyl 2-propanol trisulfide</td>
<td>1.68</td>
</tr>
<tr>
<td>3-Vinyl-1,2-dithiocyclohex-4-ene</td>
<td>47.78</td>
</tr>
<tr>
<td>3-Vinyl-1,2-dithiocyclohex-5-ene</td>
<td>15.33</td>
</tr>
<tr>
<td>Unknown&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Nd</td>
</tr>
<tr>
<td>Di-2-propenyl trisulfide (isomer)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Nd</td>
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<tr>
<td>Di-2-propenyl trisulfide</td>
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</tr>
<tr>
<td>Diallyl tetrasulfide</td>
<td>1.78</td>
</tr>
<tr>
<td>Benzenecetaldehyde</td>
<td>Nd</td>
</tr>
<tr>
<td>Total disulfide&lt;sup&gt;c&lt;/sup&gt;</td>
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</tr>
<tr>
<td>Total trisulfide&lt;sup&gt;d&lt;/sup&gt;</td>
<td>12.08</td>
</tr>
<tr>
<td>Total vinyl dithiins&lt;sup&gt;e&lt;/sup&gt;</td>
<td>63.11</td>
</tr>
<tr>
<td>Total diallyl sulfide&lt;sup&gt;f&lt;/sup&gt;</td>
<td>23.06</td>
</tr>
<tr>
<td>Total acyclic sulfides&lt;sup&gt;g&lt;/sup&gt;</td>
<td>26.40</td>
</tr>
</tbody>
</table>

CSC: supercritical CO$_2$; MEP: soaking with petroleum ether; SAE: Soxhlet with ethanol; SEP: Soxhlet with petroleum ether; MAE: soaking with ethanol; MA: soaking with water.
Nd: not detected by the equipment.

<sup>a</sup> Considered unknown because of the low probability found in the mass spectrometer;<sup>b</sup> the compound di-2-propenyl trisulfide was identified in two different retention times in the MAE extract, one of these with a probability of 95.5% and the other with 98.8%; it is assumed that this last as indicated compound and the first as an isomer given the very high probability;<sup>c</sup> Methyl 2-propanol disulfide and diallyl disulfide;<sup>d</sup> Dimetil trisulfide, methyl 2-propanol trisulfide and di-2-propenyl trisulfide;<sup>e</sup> 3-vinyl-1,2-dithiocyclohex-4-ene y 3-vinyl-1,2-dithiocyclohex-5-ene;<sup>f</sup> Diallyl disulfide, di-2-propenyl trisulfide and diallyl tetrasulfide;<sup>g</sup> Diallyl tetrasulfide, di-2-propenyl trisulfide, methyl 2-propanol trisulfide, diallyl disulfide, dimethyl trisulfide, and methyl 2-propanol disulfide.

Note: The compounds found in greater concentration in the extract are named.

**Figure 2.** Chromatogram with peaks that indicate identified compounds from petroleum ether soaking extract.
this time different compounds were obtained that were not of interest for the research, mainly long chain hydrocarbons. The main compounds of CSC extract are 2 vinyl dithiins, which together represent 63.11% (Table 3). These compounds were also identified by Yu et al. (1989), Abu-Lafi et al. (2004) and Calvo-Gomez et al. (2004). Block (1992) proposed that the conversion of allicin in vinyl dithiins, as well as in other compounds, is evident with chromatographic analysis under certain conditions. To verify that the transformation of allicin does not occur during the extractive process, Abu-Lafi et al. (2004) used cryogenic extraction techniques and CG/MS and high performance liquid chromatography (HPLC) as analysis methods and still obtained allicin as the predominant compound in all the extracts analyzed, concluding that these compounds are the major sulphurated components of fresh garlic and that during some processes (vapor distillation, frying or bleaching) they decompose and form other compounds. In the extracts obtained with petroleum ether higher relative percentage of trisulfides were found (MEP: 36.407% and SEP: 41.03%) and, in those obtained with ethyl alcohol, somewhat lower percentages were obtained (MAE: 32.743% and SAE: 30.446%); in contrast, in those obtained with water and supercritical CO$_2$ the trisulfides percentages were substantially lower (CSC: 12.08% and MA: 10.50%) (Table 3). Figure 3 shows the structures of the main compounds found.

Interaction with the solvents seems to be a very influential factor in the formation of compounds within the extracts. Notable is the case of extracts obtained using petroleum ether and ethyl alcohol, whose relative concentrations of dimethyl trisulfide range from 30.47% to 41.03%, while that in water and CO$_2$ reached 12.08% as a maximum. Cyclic compounds predominate in the latter extracts. The permanence time of CO$_2$ with the extract was the same as the extraction process (1.5 h), consequently it vaporized at room temperature, while with the other solvents the permanence time was 8 h (Soxhlet) and 24 h (maceration), plus the lapse between extraction and separation of the solvent. Iberl et al. (1990) propose that vinyl dithiins are not directly produced by catalyzed reactions by allinase, but rather by the allicin decomposition produced in solvents less polar than water.

Temperature also appears to influence the type of compounds found in the different extracts. Vinyl dithiins were found mainly in the extracts obtained at low temperatures (except for 8.19% found in one of them in the SEP extracts at approximately 60 °C). Nevertheless, a simple linear correlation between process temperatures and vinyl dithiins percentage in each extract showed that there is no statistically significant relationship among them ($r^2 = 11.7894\%$ and $p = 0.5052$). However, analyzing only the information of the extracts obtained with ethyl alcohol and petroleum ether it was established that there is a statistically significant relationship between temperature and vinyl dithiins percentage ($r^2 = 73.232\%$ and $p = 0.1212$). It is very probable that the greater toxicity of CSC extract is due to the high percentage of vinyl dithiins compared to the other extracts. As well, it was found that the CSC

![Figure 3. Structures of the main compounds in the extracts.](Source: NIST98 database (National Institute for Standards and Technology, 1998)).
extracts had very small quantities of other sulphurated compounds others than vinyl dithiins, di-2-propenyl trisulfide, methyl 2-propanol disulfide and diallyl disulfide and tetrasulfide, resulting in a relatively clean product. These observations are in accordance with Calvey et al. (1998) and Ryback et al. (2004). The aqueous extract also has a relatively important percentage of vinyl dithiins (33.52%) probably because dicroloromethane was used to obtain the extract for chromatographic analysis, a solvent that was also used to dissolve the extract obtained by supercritical CO₂ to carry out the same operations.

Another important compound in the CSC extract was diallyl disulfide (DADS), with the greatest percentage of all the extracts (10.88%). Auger et al. (2004) found that the LC₅₀ of this pure substance against R. santonensis and R. grasssei was 3400 and 2500 mg kg⁻¹, respectively; thus suggesting that DADS can also be responsible for acaricide activity. In this same study dimethyl disulfide showed a LC₅₀ against these two termités of 150 and 100 mg kg⁻¹, respectively; this compound was not found in any extraction in the present research, perhaps because of its high volatility, its capture not being possible under the conditions used in this study.

CONCLUSIONS

The garlic bulbs extract obtained with supercritical CO₂ showed the lowest CL₅₀ against T. urticae under laboratory conditions in comparison to the other extracts. The high concentration of cyclic compound could be the explanation for this effect.

There are no significant statistical differences between most of extraction methods in terms of repellence against T. urticae with the concentrations analyzed in this study (except for the aqueous extract and the CSC at 1000 and 2000 mg kg⁻¹). The concentration of acyclic compounds explains 87.21% of the repellence of each extract on T. urticae (with a 99% confidence level).

Extraction with supercritical CO₂ provided higher concentrations of sulphurated compounds, in comparison to the other techniques used in this study, resulting in a relatively clean product.

The results suggest that with extraction methods at high temperatures acyclic compounds are obtained and that processes at room temperature favor the formation of cyclic compounds in greater proportion. The extract obtained at room temperature using petroleum ether was more toxic than the other extracts (except that obtained with supercritical CO₂) and while it did not show significant differences form the other treatments, it was observed that this extract had a stronger repellent effect.

RESUMEN

Comparación y caracterización de extractos de bulbos de ajo (Allium sativum L.) y su efecto en la mortalidad y repelencia de Tetranychus urticae Koch (Acarí: Tetranychidae). Carlos Augusto Hincapié Ll.1, Gloria Eugenia López P.1, y Ricardo Torres Ch.2. Tetranychus urticae Koch es una plaga que causa grandes pérdidas económicas porque ataca un numeroso grupo de cultivos. Además, algunas poblaciones han desarrollado resistencia a acaricidas comerciales. Este trabajo compara diferentes extractos a partir de bulbos de ajo (Allium sativum L.) a través de su toxicidad y repelencia sobre T. urticae. Los extractos se obtuvieron usando como solventes CO₂ en condiciones supercríticas (CSC), etanol y éter de petróleo usando el método Soxhlet y maceración y agua usando maceración. La técnica de fluidos supercríticos permite obtener extractos a temperaturas bajas usando altas presiones, evitando la degradación de los compuestos y permitiendo su separación del solvente al exponerlo a temperatura ambiente. El ácaro fue criado en laboratorio sobre plantas de fríjol (Phaseolus vulgaris L.) en el Campus Laureles de la Universidad Pontificia Bolivariana, Medellín, Colombia. La mortalidad fue evaluada colocando 10 ácaros en un disco de hoja de P. vulgaris de 3 cm de diámetro previamente sumergido en el extracto. Las concentraciones letales medias (CL₅₀) más bajas se obtuvieron con el método CSC (8 118,8; 5 410,5; 2 820,6 mg kg⁻¹ a las 24, 48 y 72 h respectivamente). Los extractos se caracterizaron usando cromatografía de gases-espectrometría de masas (GC/MS), obteniendo en el extracto CSC principalmente vinilditiínas (63,11%), dialil disulfuro (10,88%) y di-2-propenil trisulfuro (10,4%). Se concluyó que la extracción con fluidos supercríticos permite obtener, a partir de bulbos de ajo, extractos con mayor concentración de compuestos biológicamente activos contra T. urticae, comparada con las otras técnicas usadas en este estudio.

Palabras clave: araña roja bimaculada, extractos de plantas, efecto acaricida, Liliaceae.
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