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Senecio angustifolius as the major source of pyrrolizidine alkaloid contamination of rooibos tea (*Aspalathus linearis*)

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ABSTRACT

Pyrrolizidine alkaloids (PAs) and their *N*-oxides (PANOs) were detected in commercial rooibos tea which resulted in an investigation into the source of the contamination. Field studies showed that *Senecio angustifolius* occurs as a common weed throughout the production area and that it contains high levels of the same PAs (and in the same ratios) as those found in contaminated rooibos tea. The weed superficially resembles rooibos tea plants and is easily overlooked during weeding and harvesting. Analysis of a large number of plant material samples, collected from plantations from seven regions in the production area, showed that the rooibos plant (*Aspalathus linearis*) does not produce PAs. The detection of PAs in some rooibos plant materials from fields heavily infested with *Senecio angustifolia* can be explained by the recently demonstrated process of lateral transfer of PAs.

1. Introduction

Pyrrolizidine alkaloids (PAs) and their *N*-oxides (PANOs) are amongst the most widely distributed natural toxins produced by plants and are known to be hepatotoxic to humans and animals (Bull et al., 1968; Mattocks, 1986). They may cause acute liver damage (centrilobular hepatocellular necrosis) when ingested in large amounts but animal studies have also shown that some of them are genotoxic, carcinogenic and teratogenic, as indicated in reports by the German Federal Institute for Risk Assessment (Bundesinstitut für Risikobewertung or BfR) and European Food Safety Authority (EFSA) (EFSA, 2011; BfR, 2013a). More than 350 PAs have been described from 6000 plant species belonging mainly to the families Asteraceae (tribes Senecioneae and Eupatoriae), Boraginaceae and Fabaceae (Smith and Culvenor, 1981; Hartmann and Witte, 1995; EFSA, 2011; Langel et al., 2011).

There is currently a world-wide concern about the presence of PAs in food and beverage products such as honey (Kempf et al., 2008, 2010, 2011; Bodi et al., 2014; Martinello et al., 2014) and herbal teas (BfR, 2013a; Bodi et al., 2014; EMA, 2014; Mathon et al., 2014; Shimshoni et al., 2015), but also in medicinal products (EFSA, 2011; Codex Alimentarius Commission, 2014; Allgaier and Franz, 2015) and fodders (EFSA, 2011; Codex Alimentarius Commission, 2014). Trace amounts occur in many food items but the dietary relevance of such low levels in terms of health and safety is not yet fully known (BfR, 2013a, 2013b; EMA, 2014; Allgaier and Franz, 2015).

As part of a research project, the BfR first discovered the presence of PAs in rooibos tea [*Aspalathus linearis* (Burm.f.) R.Dahlgren, family Fabaceae] and in several other herbal teas such as fennel, chamomile, peppermint and nettle, as well as in green and black tea (BfR, 2013a). Other reports of herbal tea samples, including rooibos tea, confirmed that PA contamination is a general problem, not unique to rooibos tea. Samples collected for these reports came from retail markets in Switzerland (Mathon et al., 2014), Germany (Mädge et al., 2015), Belgium (Huybrechts and Callebaut, 2015) and Israel (Shimshoni et al., 2015). The alkaloid profile given for rooibos tea showed senecionine *N*-oxide as main compound (up to ca. 300 μg/kg), followed by lower levels of senecionine, retrorsine *N*-oxide, retrorsine, seneciphylline *N*-oxide, senecivernine *N*-oxide and a small amount of senkirkine (to yield total PAs of up to ca. 500 μg/kg) (Mathon et al., 2014). In an updated report on analyses of a wide range of teas and food items (Muller et al., 2015), the EFSA found the highest mean level of PAs in rooibos tea, namely 7.99 μg/L tea infusion.

Rooibos tea is very popular in South Africa, not only because it is produced here, but also for its caffeine-free status and beneficial health properties, in particular to relieve infantile colic. Major international markets are Germany, the Netherlands, UK, USA and Japan (Joubert and De Beer, 2011). The present study was initiated to address the

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urgent need to understand the various sources of PA contamination in rooibos tea and to identify the main source of contamination. It was also necessary to understand whether the rooibos plant produce PAs, given its close relationship with the genus *Crotalaria*, a well-known source of toxic PAs.

2. Materials and methods

A survey of the entire rooibos production area was conducted in September 2014 in the Western and Northern Cape Provinces of South Africa in seven regions, including two sites in the Clanwilliam area (listed from north to south – Fig. 1): (1) Nieuwoudtville, (2) Gifberg, (3) Agter-Pakhuis, (4a, 4b) Clanwilliam, (5) Paleisheuwel, (6) Citrusdal and (7) Piketberg. All potential sources of PA contamination, growing as weeds in and around rooibos tea plantations, were photographed, recorded and collected.

2.1. Samples

Samples of PA-producing weeds were collected in rooibos tea plantations (voucher specimen numbers in parenthesis, LV = Long and Van Wyk, all in JRAU): (1) *Amsinckia menziesii* (Lehm.) A. Nelson & J.F.

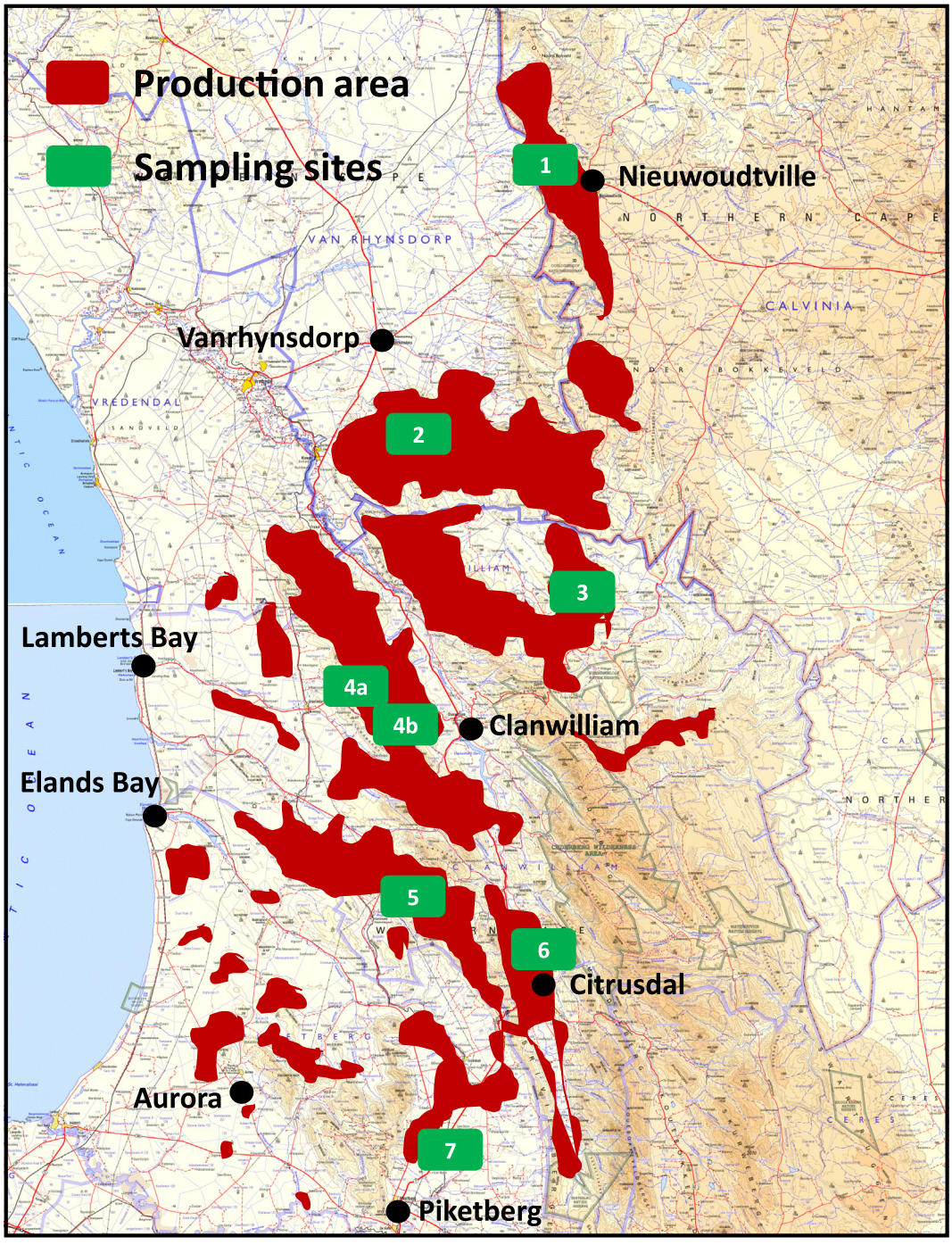

Fig. 1. Map of the rooibos tea production area in South Africa (in red) with sampling sites indicated in green. (For interpretation of the references to color in this table, the reader is referred to the web version of this article.)

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