

Cropwatch Newsletter June 2007: Alliance Special!

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Editorial Comment.

As Cropwatch continues to grow, it seems expedient to make alliances with organisations which share mutual aims & opinions. To this end, Cropwatch announces a collaboration with The Perfume Foundation in Brussels (see §7. below).

In this June 2007 Newsletter issue we also revisit a few older topics, simply because we would like to point out that some of the persisting myths amongst the aroma-interested community, are simply, just, well, persisting myths! So, in article §1 below, we say goodbye to “quenching” – although you may still see the concept perpetuated in modern aromatherapy literature, at the very least. Next (§2) we challenge the dogma of the oilseed boards, who continue to deny the existence of rapeseed allergy in spite of a mounting dossier of evidence to the contrary. We also highlight the plight of endangered plants, *Coleus forskohlii* & Candeia (*Eremanthus erythropappus*), which cosmetic & pharmaceutical companies have been so very, very busy exploiting in previous years. In the case of the Candeia plant, these self-same companies which have added to the current ecological demise of the species are currently trying to hype-up their image in the trade press by convincing trade journalists (hardly difficult!) that they are earning ‘brownie points’ by adopting taking measures to ‘save’ the species! As usual, we offer a ‘right of reply’ facility to any aggrieved parties who feel that their sensibilities have been affronted by these or other topics!

§1. Quenching: “A Cage in search of a Bird¹.”

[¹: With sincere apologies to Frank Kafka].

Background.

The term ‘quenching’ referred to a phenomenon where, apparently, it was thought that the induction of (skin) sensitizing potential of one fragrance ingredient could be nullified by the presence of another, thus rendering it safe. Opdyke (1976) had originally given examples of this process for three individual fragrance chemicals, triggered by observations with citral-containing essential oils which allegedly demonstrated lack of sensitization potential, compared with the weak to moderate sensitiser, citral.:

| Chemical: | Naturally occurs in: | Quenched by: |
|--|--|--|
| Cinnamaldehyde | Cassia, cinnamon oils etc | Eugenol 1:1 |
| Citral (= mixture of neral + geranial) | Lemongrass, litsea cubeba & citrus oils etc | d-limonene or alpha-pinene 1:4 with citral |
| Phenylacetaldehyde | Minor component of many essential oils & fruits. | Beta-phenylethyl alcohol or dipropylene glycol 1:1 |

Table 1 – Examples of sensitizing chemicals which could be appropriately quenched, according to Opdyke (1976).

IFRA subsequently re-issued Standards for these ingredients e.g. for citral (IFRA 1980) reflecting the fact that inhibition of the induction of sensitization could be prevented by addition of the appropriate materials e.g. 25% of the concentration of d-limonene in the case of citral.

Many of us concerned with the properties of natural products, have strongly and persistently maintained that the properties of individual chemicals that naturally occur in complex biological materials (such as essential oils) cannot necessarily be determined by toxicological studies conducted in isolation with commercial synthetic versions of the individual naturally occurring chemicals. One of the several arguments against this way of working is because matrix effects – the effects of co-occurring components in natural materials – are not considered. To some therefore, including many of those in the aromatherapy profession, such as Robert Tisserand (Tisserand 2003), quenching was seized upon as ‘proof’ of the fact that matrix effects could be shown to operate in complex biological materials.

Some of us were less than convinced by the quenching hypothesis, however. Firstly, and quite importantly, no robust physiological biochemical/immunological mechanism to support the hypothesis had ever been advanced (N.B. we use the word ‘robust’, because those mechanistic suggestions which have been advanced - chemical interaction, altered absorption, anti-inflammatory activity, and competitive inhibition (Guin *et al.* 1984), are all shot through with holes!). Secondly, the phenomenon was not reproduced satisfactorily, by either the original workers (who failed to properly document their original experimental conditions) or by subsequent investigators. Basketter (2000) wrote an excellent review paper on the subject entitled “Quenching: fact of fiction?”, - the broad conclusion of which is that the quenching phenomena is a hypothesis without proof. Other toxicologists have previously distanced themselves in print on this issue also. However some credence for the existence of quenching was thought to be gained from the work of Api & Isola (2000), who apparently showed that citral in conjunction with certain terpene/alcohol mixtures, inhibited the induction of skin sensitisation in humans, mimicking the conditions found in essential oils.

In spite of Basketter’s article mentioned above, Tisserand (2003) subsequently wrote a paper in support of the quenching hypothesis, citing the work of Hanau *et al.* (1983), Guin *et al.* (1984) & Allenby *et al.* (1984). Basketter (2000) had previously argued that Hanau article, at least, related to the binding of chemicals to random skin protein (soluble or insoluble), and was therefore largely irrelevant, since it was only the specific binding, (probably to surface proteins of Langerhans cells), which was important in the initiation of sensitization, after the work of Breit (1982). The earlier work of Guin *et al.* (1984) & Allenby *et al.* (1984) cited by Tisserand, has to be viewed in context with Basketter’s subsequent work with citral-limonene and cinnamaldehyde-eugenol mixtures using the guinea-pig maximization test, where no indication of quenching was found (Basketter 1991a,

Basketter 1991b). Similarly more modern LLNA studies in mice seemed to indicate a similar conclusion to Basketter's in 2000.

In the same year (2000) the SCCP adopted a Position Paper on quenching (SCCNFP/0294/00) which concluded that "Thus on the balance of the evidence presently available, the existence of quenching of certain fragrance allergens by other specific fragrance components should be regarded as a hypothesis only" – Basketter's opinions & words more or less exactly (Basketter 2000).

Four years on, Lalko & Api (2004) of RIFM published a paper entitled "The potency of citral in the Local Lymph Node Assay" which considered the sensitizing potential of citral plus two citral-rich essential oils (Lemongrass oil & Litsea cubeba oil) applied in a 1:3 ethanol: DEP (!) vehicle (*Cropwatch comments*: deployment of DEP in this testing regime is surely a curious choice given the strong anti-phthalate feelings amongst 'green' cosmetics consumers worldwide).

The authors found EC3 values (EC3 values are said to represent relative potency according to the methodology of Basketter *et al* 2000b) of 6.5% and 8.4% for Lemongrass & Litsea cubeba oils respectively, which was not significantly different from that for citral (6.3%). They further found little significant difference between citral quenched with limonene & straight citral (EC values of 0.8% & 1.2% respectively). Further, the authors found that in humans a NOEL of ~0.5% or 1400µg/cm² exists for the induction of sensitization to citral. In conclusion, Lalko & Api conceded that Basketter (2000) had shown that quenching could not be demonstrated in conventional animal models such as the guinea-pig maximisation test, & concluded that the quenching effect of citral could not be demonstrated in the LLNA assay either.

Where this puts the credibility of the previous Opdyke (1976) & Api & Isola (2000) findings, where the induction of sensitization of citral-containing essential oils or citral-terpene or terpene alcohol mixtures was shown to be obviated, is not clear to us at present – presumably either these findings are to be ignored, or the interpretation of the results from earlier animal models & LLNA test has shortcomings which we need to know about. Either way, we don't seem to have been offered an explanation, and with so much credence apparently resting on the LLNA testing procedure in corporate toxicologist-led sensitization studies, Cropwatch feels that this matter should be cleared up.

Further, this incomplete study only seems to beg further questions – citral is the term describing a chemical mixture of structural isomers of neral & geranial in various ratios according to origin. Any effects due to isomer distribution were seemingly not evaluated or discounted. The purity of citral used was only 99.5% - effects of impurities were not mentioned. The species & geographic origins of the Lemongrass oil were not stipulated (Lemongrass oils from *C. flexuosus* Nees ex Stued., *C. citratus* (DC) Stapf. and *C. pendulus* Nees ex Stued. are all easily

commercially available). Likewise, in practice, commercial 'Litsea cubeba oil' is distilled from several species apart from *Litsea cubeba* including *Litsea enosma* & *L. mollifolia*. Further, commercial Lemongrass & *Litsea cubeba* oils frequently contain adulterants like synthetic citral, and blind trust on industry to provide authentic oils 100% derived from the named botanical species is simply not professionally acceptable. Best practice dictates that the essential oil plant-source has to be identified at source by a suitably experienced botanist, and the derived oil batch subsequently lot-tracked to guard against interference (adulteration), or, that the investigators personally distill their own test material, as of course, many workers do. Finally, we have seen criticisms supporting the contention that the LLNA method is unreliable with weak sensitizers (presumably such as citral) and the results in this area can be open to interpretation. For industry to have any faith in this methodology, we need to understand these issues more completely.

IFRA Ducks Out.

In June 2004, according to the IFRA Hazards Working Group opinion "quenching phenomena" effects can still be taken into account (according to Section 3.3 of the EU Dangerous Preparations Directive 88/379/EEC); however quenching phenomena effects between eugenol and cinnamic aldehyde are now unsupported according to the Notification No 4. of 38th Amendment to the IFRA Standard."

On to more immediate times, the end-note of the revised citral standard under the IFRA's 40th Amendment 2007 quietly says (not of course, any apology to industry for the fact that their previous guidance may have been wrong!) but:

"This Standard cancels and replaces the existing one on citral, which was based on the no longer supported 'quenching' phenomenon."

So, in spite of all the previous confused messages, in 2007 (apparently) IFRA is indicating that it no longer supports quenching, period. You still may find this news is a little surprising, revealed as it is, buried at the bottom of a document. After all, it is only 3 years ago since Lalko & Api clearly stated "The quenching of sensitization to citral by the addition of d-limonene has been demonstrated in humans" (Lalko & Api 2004), and It is only 1 year ago (actually March 2006) since Api and her aroma-industry toxicology pals (including Basketter), reproduced the fact that under the existing IFRA Standard, citral has no concentration limit wrt their given example (hydroalcoholic products for unshaven skin, and in solid antiperspirant product types) (Api *et al.* 2006).

30 Years of Argument Ended over the Existence of a Quenching Effect.

So there you have it. Apparently we have to write off the findings a lot of previous published dermatological work (Nilsson *et al.* 2004, Safford *et al.* 1990, Guin *et al.* 1984, Allenby *et al.* 1984, Hanau *et al.* 1983, Api & Isola 2000 etc.) which seemingly helped to confirm the existence of a quenching effect. If we now

accept this work as being flawed, it must beg many questions about professional standards of published research work in dermatology.

N.B. Cropwatch does not support animal testing of cosmetic ingredients in any shape or form, including the LLNA assay procedure, but merely reports above on data publicly available in the scientific literature.

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§2. Rapeseed (Canola): Allergic Properties Revisited, & Wrongful Thinking on its Potential as a Biofuel.

(Modified from the original article "Rapeseed (syn. Canola) Revisited" posted to the Aromaconnection blogsite by Tony Burfield on 21st April 2007 at www.aromaconnection.org)

For a few weeks in Springtime, parts of England's 'green and pleasant land' turns into a nauseous Day-Glo Yellow vista, as farmers submit us hapless country-lovers not only to hideous horizon-wide visual onslaught, but also to our annual toxic gassing from the sweet, cloying but underlyingly unpleasant volatiles from rapeseed flowers (*Brassica napus* L. ssp *oleifera*) coupled with a seasonal overload of rapeseed pollen. Although plant breeders have overcome the toxic nature of the original plant to some extent – which originally contained high levels of aliphatic fatty acids, such as eicosenoic and (more especially) erucic acids - the high pollen levels, the high levels of insect-attracting nectar (especially hoverflies), and the disgusting volatile sulphur producing properties, are still present.

Honeybees (*Apis mellifera* L.) are apparently attracted mainly by the Rapeseed's flower's volatiles: particularly phenylacetaldehyde, linalool & (*E,E*)-farnesene (Blight *et al.* 1997), and the cabbage seed weevil *Ceutorhynchus assimilis* is attracted to its host plant, *Brassica napus*, by volatile isothiocyanates (Bartlet *et al.*, 1993), perhaps including 3-butenyl, 4-pentenyl, and 2-phenylethyl isothiocyanates during certain development cyclic stages (Smart & Blight 2004). However several of us mere humanoids might react more adversely to unpleasant substances such as the phenylacetone nitrile, isothiocyanates & the respiratory irritant dimethyl sulphide (McEwan & MacFarlane Smith 1998).

According to the summary provided in the article introduction by Smolinska *et al* (1997) and the references therein, in tissues of the Brassicaceae, it appears that after tissue damage, myrosinase (thioglucoside glucohydrolase) hydrolyses the naturally occurring anionic glucosinolates present (which consist of beta-D-thioglucose and sulphonated oxime moieties), producing a number of end products including isothiocyanates, thiocyanate ion, nitriles, and epithionitriles according to the type of glucosinolates present and the exact hydrolysis conditions. As insect feeding deterrents, these products of myrosinase activity are more toxic than the glucosinolates themselves (Eckardt 2001).

A search of the available literature has not revealed any detail whether the aroma volatiles produced by the rapeseed flowers involve similar processes in generating for volatile sulphide compounds.

Uses - bio-diesel & cosmetics.

When the UK crop matures, some of the fixed oil produced is sold for bio-diesel, mainly to Germany, according to an illustrated article in a recent Guardian supplement (Blythman 2007). Elsewhere, it is quoted that some 40-60% of European rapeseed production goes to bio-diesel production (Mudeva 2006); Apart from France, Poland is also among top rapeseed-oil producing countries, its' output being quoted as 1.49 million tons in 2004 (Krukowska 2004), although UK production is expected to top 2 million tons in 2008 (Blythman 2007). DEFRA have previously pointed out that 1 ton of rapeseed gives 0.38 tons of rape methyl ester (one possible bio-fuel, besides rape ethyl ester etc.). In other parts of the world, opposition to bio-fuels, especially bio-ethanol, has been vigorous (see Larouchepac 2006), but it hasn't stopped the Canadian Agriculture minister in 2006 announcing \$11 million in funding initiatives for Canadian farmers in bio-fuels opportunities (N.B. already 40% of the rapeseed grown in Canada is GM, according to Teitel 2001).

Supplementing diesel is apparently in line with the EU commitment to increasing the share of bio-fuelled transport to 10% (Kroeger, 2007), although this has bought criticism from many environmental NGO's who say the policy will do more harm than good (Anon 2007). Some of these criticisms have been aired in the UK national press, such as the fact that land, which should be primarily set aside for food production, is being raffled off to appease the modern great God: the motor car. In a slightly different area, George Monbiot has criticized the UK government for not disqualifying palm-oil (which he maintains will actually worsen greenhouse gas discharges) from EU-driven bio-fuel targets (Monbiot 2006). True, a public consultation exercise on the bio-fuels policy topic was conducted by the EC (EC 2006), but as usual, the EC was more concerned with policy than practicality, and failed to ask views on the pertinent issues.

In Canada, low erucic acid (< 2%), low glucosinolate (<30 micromoles/g air-dried oil-free meal) GM rapeseed bred for herbicide resistance is called Canola (Canada – oil). Resistance to GM canola cropping seems to be lower in Canada

– elsewhere worries about corn contamination from GM material & associated public liability issues, quite apart from high consumer resistance, have been reported (GMWatch 2004). However pro-GM lobbyists continue to exert pressure in countries like Australia (NCF 2006).

Low erucic acid rapeseed oil is used in cooking & margarine production, and in the preparation of salad dressings. The oil is used as a lubricant as well as an ingredient for bio-fuels as discussed; the oil free press-cake is high in protein and is used in the animal feed industry. A small amount of rapeseed oil is used in cosmetics, especially in soap-making, as a carrier for fragrances in 100% 'natural' perfumes, and as a diluent for candle fragrances, in lamp oils etc. However, even if from a non-GM source, considering rapeseed oil's negative eco-associations as set out below (nitrate leaching, crop spraying requirements etc.), any usage of the oil as a fragrance diluent is restricted to the 'natural' perfumes category, and for obvious reasons claims cannot be extended to 'organic' perfumes.

Rapeseed oil production – the negative aspects.

Although rapeseed *Brassica napus* L. is native to Europe and has been cultivated since Neolithic times, committing a large area of agricultural land to growing rapeseed is a new development, and has a downside. Some of the issues here include:

1. The increased use of harmful crop sprays. Rapeseed is prone to widespread attack from a variety of insect & microbiological predators. An average crop might receive the following sprayings: 3 of herbicides, 2 of fungicides & 2 of insecticides per growing season (Office of National Statistics through Blythman 2007). Insecticides commonly used include glufosinate ammonium & the hormone disruptor vinclozolin.

2. Possible eco-damage. Rapeseed crop production is associated with higher demands for nitrogen & sulphur-based fertiliser application, and excessive nitrate leaching into water sources is associated with rapeseed cultivation, causing localised environmental problems. In addition, decaying rapeseed vegetation (in common with other *Brassica* spp.) is known to put thiocyanate into the soil (Brown & Morra 1993), & soils treated with defatted rapeseed meal were determined to yield 6µg/g of thiocyanate (Brown *et al* 1991), although please note that a determined chemical value for thiocyanate, & and its total bioavailability, may differ. Microbiological degradation over several days will offer the principle detoxification route for thiocyanate (Brown & Morra 1993).

However the Canadian Canola Board indicates that breeders of Canola varieties have reduced the glucosinolate contents in rapeseed meal (where the bitter taste of glucosinolates acts as a feeding deterrent). Already they claim Canadian Canola meal has only an average 16µg/mol total glucosinolates (and some years it has been lower than this), compared with traditional meal which contains 120-

160µg/mol. Judging by the nitrile or isothiocyanate volatiles coming from flowering rapeseed fields in the UK, this sort of technology hasn't yet spared UK citizens from their annual gassing.

3. The GM issue. Monsanto has been amongst those companies producing transgenic rapeseed varieties, modified to be resistant to RoundUp (known in Australia as RoundUp Ready canola). You may remember Monsanto previously hit the headlines when it prosecuted a Canadian farmer, who, it was claimed had allegedly infringed their property rights (wind-blown (?) GM rapeseed plants had appeared on his land). The judge, to the outrage of GM protesters, found for Monsanto (Teitel 2001), and although the case was later reviewed, the court still found for Monsanto (BBC 2004). The EC halted new approvals for GMO's in 1998 due to intense consumer opposition, but the US filed a complaint at the WTO in 2003, supported by Argentina & Canada. The US action has been widely seen by EU consumers as bullying, and since the FDA has been involved in international GMO promotion, Cropwatch now sees this organization not as a reliable independent health authority, but as an authority hopelessly tainted by political influence.

In 2004 the EU introduced labeling & traceability procedures for GMO's, but meanwhile the US has proved to be the leading source of global GM contamination. For example between 2001 & 2004 hundreds of tons of maize contaminated with Sygenta's unapproved transgenic variety Bt10 were distributed world-wide and entered the global food chain, without the US authorities noticing for these four years...

The EC authorised Monsanto in August 2005, in a totally undemocratic move, to be allowed to grow the GM rapeseed variety GT73 in Europe for 10 years, going against the wishes of the EU member states, 13 out of 25 of whom had voted against the proposal. The EU Commissioners seem, for unknown reasons, keen to promote GM technology throughout Europe, and are out of touch with the opinions of the majority of EU citizens who maintain a strong anti-GM stance. A more recent evaluation of the safety of GM canola, including the explaining away of increase in liver weights of rats fed GT73 canola, is to be found at FD Govt Au (2007).

The European Commission just recently authorized the Bayer Chemical Company to be allowed grow three GM rapeseed varieties in Europe for the next 10 years, modified to resist glufosinate ammonium. And so it goes on.... the financial might & influence of big industry wins out against the wishes of the people.

4. Allergic Reactions

According to my unscientifically-based observations (i.e. talking to some UK GP's), rapeseed pollen causes untold seasonal respiratory misery for a proportion of the (rural?) UK population, but this fact is apparently disputed by

oilseed organizations. The scientific press shows little clear direction on the issue either – just a handful of articles, both for (e.g. Focke *et al.* 1998; Hemmer *et al.* 1997) and against (e.g. Gylling 2006) an allergic association. Previously Parrat *et al.* (1995) had shown in a Scottish study that allergic reactions were not directly related to airborne pollen levels, although Welch *et al.* (2000) ruled out cross-reactivity with grass pollen. Soutar *et al.* (1994) investigating 1000 people from the Aberdeen area had suggested the prevalence of symptoms was small and could be caused by chemicals from the crop chemicals. Similarly Murphy (1999) had concluded that rapeseed allergenicity only had a minimal impact on health. An article by Butcher *et al.* (1994) looked for possible aeroallergens/irritants & identified 22 volatiles from rapeseed flowers.

More recent studies paint a more illuminating picture, however. Children with IgE-mediated allergy to foods often show reactions to rape seeds in skin prick tests (pathways unknown). Puumalainen *et al.* (2005) have shown that 2S albumins (seed storage proteins) may be responsible for the rapeseed food allergy, and investigations characterising these proteins being investigated by Palomeres *et al.* (2002). More recently, Fiorina *et al.* (2003) employed an *in situ* aerobiologic test to detect the presence of a rapeseed allergen, where routine tests had failed, and Hermanides *et al.* (2006) describe cases of occupational allergy to *Brassica* pollens.

It seems then that science has yet to recognize the UK people's anecdotal experiences of eye & upper respiratory irritation from rapeseed volatiles or pollen and offer some proper explanations. Or is there a conspiracy of silence? An excellent thread from 1996 on the Gentech archive (Gentech 1996) shows examples of academic unawareness (failing to find what published studies there are), UK ministerial indifference (no evidence, but this is revealed to be an empty defence which simply reflects a lack of authoritative studies) and a surprising dearth of North American & Canadian anecdotal symptom reporting, in contrast to the UK experience. The Gentech article also provides 14 references related to allergenicity of rapeseed/rapeseed products – which had proved so hard for some authorities to find (err, no change there then!).

Concluding Remarks.

The persistence of public belief that rapeseed cultivation causes widespread seasonal respiratory distress has been remarked upon by Blythman (2007), who maintains (paraphrasing his words) that in the absence of a clear case of causation, maybe we should own up to the fact that in the UK, we simply don't like the stuff. I, for one, am quite willing to own up to that fact. However it is more likely that better investigative science will conclusively reveal a causative mechanism for the allergy syndrome, forcing a reversal of the current 'in-denial' attitudes of oilseed boards, plant breeders & agricultural officials.

Postscript. After this article was largely complete, we discovered a website devoted to the rapeseed allergy syndrome - **please visit it at** <http://www.oilseedrape.org.uk/>

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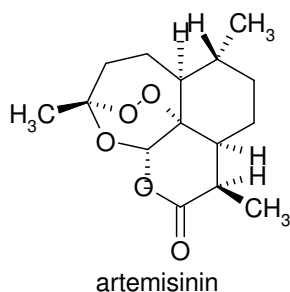
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§3. CNAP Artemisinin Project Update.

Cropwatch has previously featured news concerning *Artemisia annua*, the herbal source of the anti-malarial drug, artemisinin. ACT's (artemisinin combination therapies) are the favoured therapeutic choice for malaria treatment, in the light of increasing drug resistance to single-active synthetic drugs such as chloroquine.



The first update from the CNAP-Artemisinin project (Spring 2007), which aims to use fast-track breeding technologies to create, non-GM *Artemisia* cultivars with increased artemisinin yields, is available at <http://www.york.ac.uk/org/cnap/artemisiaproject/index.htm>. The project centered at York University has reportedly joined forces with the Medicines for Malaria Venture and the Institute of One World Health, coordinating efforts to secure affordable supplies of artemisinin as the *Artemisinin Consortium*.

§4. Threatened Species: *Coleus forskohlii* revisited.

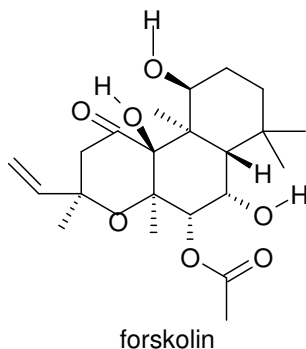
At Cropwatch, we always get nervous when we read that large corporations take out patents centered around the useful properties of rare or endangered species, or are reported as marketing significant amounts of herbal extracts from these same ecologically-pressurised plants. This is because however correctly these companies may or may not behave, they help to create a situation of increased demand for already beleaguered species, leading to the prospect of their extinction in the wild. Our nervousness is not lessened by our perceptions of the anti-environmental/pro-industry stance of officialdom e.g. in the shape of the EU Cosmetics Commissioner, who, when eventually replying (31.10.06) to Cropwatch's submission for a list of corrections to the EU Cosmetics Inventory, disagreed with us that rare & threatened species should be removed from the Inventory, and stated that industry was free to use them. And use them it does, in some cases driving useful species to the edge of extinction (cf. Kenyan Cedarwood), sometimes out of profound ecological ignorance, but more often, we suspect, simply because profit is more important than any sustainability considerations.

In the very first Cropwatch Newsletter (May 2004) we reported the fact that the Sabinsa Corporation had patented a method (US Patent # 6,607,712) to optimize the isolation of an anti-microbial essential oil via the supercritical CO₂ extraction of the roots of indigenous & rare *Coleus forskohlii* Brig (syn. *C. barbatus* Benth) plants, which they claim can be used against cutaneous infections (the oil is said to be active against *Propionibacterium acnes*), and in combating the growth of *Streptococcus mutans* (important in tooth decay). Burfield (2007) has previously been sent unsolicited samples of deep rusty red liquid samples of steam distilled *Coleus forskohlii* root essential oil, which he describes as having a muted, slightly smoky woody odour (reminiscent of burnt timbers), with waxy somewhat green

and aldehydic notes also present. Misra *et al.* (1994) described the major constituents of the root oil from Indian plants as containing 3-decanone (7.0%), bornyl acetate (15%), β -sesquiphellandrene and γ -eudesmol (12.5%) plus and unknown sesquiterpene hydrocarbon (7.5%).

This same plant, which has edible roots & is indigenous to Indian Ayurvedic medicine (Ammon & Kemper 1982), has been indiscriminately gathered from the Indian plains and hill slopes; but it is also distributed throughout Nepal, Burma & Thailand. In India at least, the plant is threatened, so much so, that researchers such as Misra *et al.* (1994) to their great credit, refer to some precautions that authors took on their own initiative to attempt to preserve the species - a commendable act. Rajasekharan *et al.* (2005) have described an *in vitro* method of conservation of *Coleus forskohlii*, as had Sharma *et al.* (1990) previously, describing an *in vitro* clonal multiplication followed by micropropagation procedure. Vieira (1999) in an article reviewing the conservation of medicinal & aromatic plants in Brazil notes that chemotypes of *Coleus barbatus* (syn. *Coleus forskohlii*), which probably originated as African imports, are cultivated by clonal propagation in Brazil.

The roots of *Coleus forskohlii* are known to be a source of the labdane diterpene substance forskolin (syn. coleonol) or 11-oxomanoyl oxide (de Souza 1991) which is a potent stimulant (adenyl cyclase activator). Saleem *et al.* (2006) have described a simple active carbon column chromatography method for the isolation of forskolin at 96.9% purity from the roots of *Coleus forskohlii*. Mukherjee *et al.* (2003) previously described increased yields of forskolin from genetically transformed cultures of *Coleus forskohlii*.



Uses.

Forskolin has been fairly extensively researched for its potential as a drug for thyroid disorders (Haye *et al.* 1985), glaucoma (Badian *et al.* 1984; Meyer *et al.* 1987), asthma (Lichey *et al.* 1984), congestive heart failures (Kramer *et al.* 1987), vascular smooth muscle relaxant (Kramer *et al.* 1987; Schlepper *et al.* 1989) and to treat certain cancers (Valdes *et al.* 1987). Richardson (1992) noted the interest in forskolin due to its cardiovascular activity and later its apparent ability to activate adenylate cyclase (Seamom & Daly 1981), but considered that claims that the plant would enter ranks of other famous plants with

cardiovascular activity as exaggerated. However semi-synthetic analogues of forskolin have been developed and may do better, especially in the treatment of glaucoma (de Souza 1993). Nevertheless, whatever the negative connotations, the potential for India to earn foreign currency from exporting herbal medicinal drugs such as *Coleus forskohlii* is very important, and the cultivation of *Coleus forskohlii* as a medical crop has been described by Shah & Kalakoti (1996). Cultivation has been reported elsewhere in India in Tamil Nadu, Gujarat & Karnataka; however the scale on which this is carried out appears very minor in Karnataka at least, from our reading of official reports (e.g. Gadgil 2004).

More recently Prakash & Majeed (2007) of the Sabinsa Corporation published a review article on phytonutrients where orally consumed nutritional substances (nutricosmetics) coupled with topically applied cosmaceuticals combine in harmony to promote physical appearance & well-being (allegedly). Inevitably *Coleus forskohlii* root extracts are mentioned, both topically as a skin conditioning agent and internally as a 10% extract for weight management. Previously Badmaev (Sabinsa Corporation) had issued a company report on forskolin usage in body weight management (Badmaev 2000). Elsewhere it is claimed that the activation of adenylate cyclase and hence cAMP then promotes the activation of hormone sensitive lipase which helps build lean body weight at the expense of body fat. In fact forskolin for promoting lean body mass & treating mood disorders, which is the subject of a US patent (# 5,804,596) - on behalf of the Sabinsa Corporation. Sabinsa's ForsLean® product for this latter weight management purpose is reported to be shifting large amounts of *Coleus forskohlii* extract in Europe (Patton 2005).

We predict that eventually *Coleus forskohlii* will become extinct in the wild in a number of existing habitats, following a path already pre-set for Amyris, Rosewood & East Indian Sandalwood – all formerly common commodities which are becoming increasingly difficult to obtain unadulterated, and in any quantity, because of their unrelenting over-exploitation by industry.

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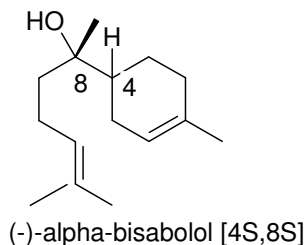
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§5. Threatened Species Cont'd: The Brazilian Candelia Plant (*Eremanthus erythropappus* (DC) MacLeish).

The aroma-giant Symrise (formed in 2003 by the amalgamation of Dragoco & Haarman & Reimer, both situated in Holzminden, in the Lower Saxony area of Germany) has announced in a sparkling piece of publicity that it is taking measures to protect the Candela plant (Prance 2007), which grows in the Atlantic Brazilian rainforest, mainly south of Minas Gerais State. Ironically, these measures come after years of the plants over-exploitation as a primary source of

(-)-alpha-bisabolol, primarily by the German pharmaceutical industry (Lopes *et al.* 1991). Previously Lauterbach *et al.* (1992) of BASF, Germany, were granted a US patent for the purification of alpha-bisabolol from Candeia extract using a reduced pressure distillation process and lower distillation column outlet.

Symrise is also amongst those to have also launched cosmetic products containing (-)-alpha-bisabolol. The reason for these years of progressive exploitation of the species has been to utilize the recognized skin healing & anti-inflammatory attributes of isolated alpha-bisabolol (e.g. see Jellineck 1984).



Alpha-bisabolol's occurrence is however not just confined to the Candeia plant – it also occurs in German Chamomile. *Chamomilla recutita* (L.) Rauschert, Horehound oil *Marrubium vulgare* L., Phoebe oil *Phoebe porosa* Mez. etc. (Burfield 2007). Schultz (2003) of Dragoco discusses the fact that for pharmaceutical and cosmetic products, chamomile essential oils with the highest alpha-bisabolol content and a very low content of bisabolol oxides are preferred. In a further article (Braun *et al.* 2003) we are told that the known sources of (-)-alpha-bisabolol are *Chamomilla recutita*, *Vanillosmopsis erythroppus* (syn. *Eremanthus erythropappus* & *Vanillosmopsis arborea* (for *V. arborea* e.o. composition - see Creivero *et al.* 1996), but that commercially, (-)-alpha-bisabolol is only isolated from the steam distilled wood oil of *Vanillosmopsis erythroppus* (Carle 1996). where it occurs at 0.1%, or more rarely at up to 0.5%. Isolating alpha-bisabolol seems to be a high carbon footprint process considering that, for example, Curado *et al.* (2006) report that the folk-medicinal Brazilian plant *Lychnophora ericoides* contains up to 76.4% alpha-bisabolol.

Prance (2007) indicates that Symrise will be working with other Brazilian partnership firms to help support & finance a project conducted by the Universidade Federal de Lavras, which aims to optimise growing conditions for the plant (*Cropwatch comments*: Lets hope this University-Industrial alliance doesn't become another disaster like the Rosewood oil cultivation situation). The above institution has already published a study on a management system for native Candeia forest (Scolforo *et al.* 2004a), and oil contents of Candeia wood, branches & leaves (Scolforo *et al.* 2004 b). In another forest management study, Prerez *et al.* (2004) have already indicated that the commercial exploitation of Candeia for fenceposts, and essential oil by-product is only feasible where the predominance of Candeia spp. constitute >70% of the vegetation.

In the applications area, interesting research on a synergistic effect (as estimated by time-kill data), between ampicillin & the essential oil of *Eremanthus erythropappus* or beta-bisabolol contained within the oil, against *Staphylococcus aureus*, has been published by Nascimento *et al.* (2007). It was also found that both the essential oil & beta-bisabolol have the potential to restore the effectiveness of ampicillin against resistant *S. aureus*.

Technical notes:

1. MacLeish (1987) discusses the revision of the *Eremanthus* noting that taxonomic distinctions between *Eremanthus* & *Vanillosmopsis* are largely artificial. Therefore the Candeia plant can have several botanical namings.

2. The chemical variability of *Eremanthus erythropappus* has been examined by Lopes *et al.* 1991. The authors interpreted occurrence of aberrant bisabolol-poor species as "a case of micromolecular diversity of sympatric species." ...err....absolutely!

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§6. Citrus Oils and Furanocoumarins; Update.

Cropwatch has been invited to a meeting with the European Cosmetics Commissioner on the furanocoumarins in cosmetic ingredients issue on July 3rd 2007 in Brussels.

In a separate development Cropwatch understands that RIFM contracted Prof. David Kirkland of Covance Ltd. UK to carry out studies on phototoxicity studies on isopimpinellin & bergamottin on their behalf, using samples obtained from Extrasynthese (RIFM 2007), and. presumably this work has been submitted by RIFM to the SCCP by the end of March 2007 deadline. We would be negligent if we did not point out that the New Jersey based-Covance animal testing company has been the subject of video-evidenced animal cruelty allegations by PeTA in their Vienna, Va. establishment (see http://blog.peta.org/archives/2007/04/covance_payspe.php), and similarly by BUAV in their investigations into operations at Muster, Germany in 2004 (see <http://www.buzzle.com/editorials/1-17-2004-49549.asp>). Altogether, we regard Covance as a pretty strange choice of a research partner by RIFM, the latter being an organisation representing an industry which should be particularly sensitive about its public image wrt the use of animals in ingredient safety testing.

Although IFRA/RIFM officials never answer our mail enquiries, when Cropwatch contacted Prof. Kirkland of Covance UK for details of the phototoxicity contracted work, he was gracious enough to explain why he had to decline, even

though we had politely pointed out this was a public interest health & safety matter. The RIFM scientific report indicates that Kirkland found that bergamottin provided a photoclastic response (structural chromosome aberrations) significantly above background at all concentrations tested in the presence of UV light, whereas isopimpinellin induced aberrations not significantly different from the controls. Unless RIFM allow Kirkland to publish the work, or the SCCP publishes the entire work itself, then presumably the exact details of this work will remain hidden from public scrutiny.

Whilst deliberations on isopimpinellin & bergamottin continue on behind closed doors, Cropwatch has been seeking industry's opinions on the furanocoumarins issue. Our broad findings are as follows, which we make no apologies for expressing in the plainest of terms.

1. The perfumery industry needs the citrus industry more than the citrus industry needs the perfumery industry (on the principle that citrus oils are by-products of the beverage industry).
2. A number of major players amongst aroma ingredient producers already have the technology to reduce or partially reduce FCF levels in citrus oils for the flavourings market, and are quietly happy to have a market advantage over the smaller concerns which haven't been able to afford the investment in the required technology.
3. These major players (see 2 above) appear to dominate policy (& research) within the major essential oil organizations.
4. These same major players (see 2 above) are, in principle, happy to provide FCF level data for their citrus ingredients to their customers, but not, in general, to impart this information to the public domain.
5. Our perception of supplier's attitudes is that they feel that customers (i.e. perfumers & technical formulators) have not created a 'demand climate' for ingredient FCF data.

As you can see we have found a situation dominated by the self-interests of big industry, where there is a real probability of financial discrimination against smaller industries if legislation restricting FCF levels in cosmetic ingredients is passed by EU legislators. The views of small producers are unrepresented, and, as usual, nowhere in this equation are socio-economic or ecological factors addressed by either industry or EU regulatory officials, who are tunnel-visioned on the 'safety' aspects only.

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§7. Collaboration between The Perfume Foundation & Cropwatch.

Pre-amble

A collaboration is announced to pursue mutual objectives for the further promotion of the Perfumery Art is announced between The Perfume Foundation & Cropwatch.

The Perfume Foundation has been operating in Brussels since 1995, and states that its mission is “to be the leading authority on environmental and health issues related to fragrances and scents, while contributing to the cultural heritage of perfume.”

Cropwatch really commenced in earnest from 2004 as an Independent Watchdog to the Aroma trade, initially to try to counter the commercial over-exploitation & bio-piracy of rare & threatened natural aromatic plants. More recently Cropwatch has tried to counter regulatory threats against the sustainability of natural product usage in everyday life.

Mutual Interest Projects:

1. To assemble, facilitate & expand the exchange of scientific information on fragrance matters.
2. To achieve a critical mass of experts in order to challenge some of the wrong-headed outcomes from deliberations of existing governmental legislators.
3. To work to launch the “Campaign for Real Perfumes” Underlying this is the belief that natural ingredients should not be unduly discriminated against by legislators. The scope and details of this campaign are still being discussed.
4. To promote agro-commerce in safe natural aromatic materials for fragrance production in order to facilitate artisan perfumery. This may include the support of producers in undeveloped nations who are discriminated against by legislation i.e. legislation which will only allow usage of ingredients which have been subject to high technology (& therefore high investment cost) processing. .
5. To create “The Perfume Embassy” office in Brussels as an ambassador of perfume excellence.

The Problems to Overcome.

Fragrance regulation in the EU unfortunately occurs within the cosmetics sector, rather than being subject to its own Fragrance Commissioner and fragrance expert advisers. Anyone working within the cosmetics sector will be totally aware that fragrance expertise is separate and non-complementary to cosmetics expertise.

Because of the way the administration has been set up within the EU, a number of critical mistakes have been made in the way the Cosmetics regulatory process operates. These mistakes need to be properly identified and put right.

Proposed Events.

It is proposed that a conference be held in Brussels at a date to be announced. The conference would feature speakers with views at variance with those expressed by IFRA/RIFM, the SCCP etc. etc. The conference could also include natural ingredient manufacturers adversely affected by cosmetics sector legislation, as well as strongly featuring the cosmetic industry & its problems. Interplay with other industries who are also opposed to EU policies in their own fields is also being sought, to see if lessons can be learned.

§8. Spice Oils & Oleoresins in Foodstuffs Under Threat.

At Cropwatch we had believed – oh so naively, it appears – that it wasn't within the remit of EU regulators, however authoritarian, to pass legislation which will effectively destroy the industries they are regulating. After reading Brussels insistent proposals to tag and monitor the movement sheep & goats in the UK – the costs of which will easily bankrupt almost all UK sheep farmers according to DEFRA (Anon 2007), we have now modified our views. But now for news of something potentially worse...

A proposal which will come before the European Parliament on 9th July 2007 under a new food law to regulate food flavourings & food ingredients (draft European Parliament Legislative Resolution Proposal report dated 15.05.07) includes Amendment 20 to Article 4 B.B, **that only spices as such should be added to food unless “there is a reasonable technological need and the desired effect cannot be achieved by using spices”**

–see

<http://www.europarl.europa.eu/sides/getDoc.do?pubRef=-//EP//NONSGML+REPORT+A6-2007-185+0+DOC+PDF+V0//EN&language=EN>

(N.B. there are some reports of trouble with this link – please apply to Cropwatch for document in case of difficulty).

This creates an impossible situation for using spice oils/spice CO₂ extracts/spice oleoresins in foodstuffs, and an industry source has verified the fact (after taking legal advice) that **“this amendment will destroy the spice oil/oleoresin business”**. It will also, of course, cause disruption at attempts to further international harmonization as many spice oils & oleoresins are official in the FCC. Perhaps most vividly, it illustrates the awareness vacuum that some EU legislators apparently operate in, where the socio-economic outcomes from their actions are simply not considered – many communities in Africa, Indonesia/SE Asia for example are economically dependent on the added value that distilled

spice oils can bring and bringing the spice industry to its knees (which this bill will do) is surely not in the spirit of world trading objectives & helping under-developed nations.

The first reading of the bill has been moved forward to the 9th July 2007 and activists are endeavouring to persuade the rapporteur, Mojca Drčar Murko to either cancel the rogue amendment (40 MEP's are needed to do this) or replace it with one which will not threaten the spice oil industry. Although some parts of industry are confident that they can muster 40 MEP's to force an amendment, their track record for getting their own way in the EU Parliament isn't 100% - but maybe we shouldn't revisit past failures on this occasion. .

Reference.

Anon (2007) "Down on the farm" *Private Eye* No 1186, 8 June-21 June 2007 p8.

Copy & paste the following links to your browser:

voting ENVI:

<http://www.europarl.europa.eu/sides/getDoc.do?pubRef=-//EP//TEXT+REPORT+A6-2007-0185+0+DOC+XML+V0//EN&language=EN#title3>

European Parliament - May 15: 2007

<http://www.europarl.europa.eu/activities/expert/committees/reports.do;jsessionid=4657F3F1BA2DF1EDD7E080D3BFD956C4.node1?committee=2353&rewritten&language=EN>

Parliamentary processes:

<http://register.consilium.europa.eu/pdf/en/07/st09/st09536-ad04.en07.pdf>

Parliamentary processes:

http://ec.europa.eu/prelex/detail_dossier_real.cfm?CL=en&DosId=194540

§9. Endangered Animal Trade Update – DNA Kit

A DNA test kit, to spot whether goods have been prepared from products from threatened species, was unveiled at the 14th Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) meeting in the Hague on June 12th 2007. The kit was developed by the WSPA & Wildlife DNA Services, a company based in N. Wales, and it is hoped that might prove especially useful against helping prevent the illegal trade in bear bile & bear gall-bladders from bear farms in China, Korea & Vietnam. The test kit has been validated for bear proteins, and is on trial in Australian Customs Service & Wildlife Enforcement Directorate, Environment Canada.

As we discussed in our review article on Threatened species & TCM products <http://www.cropwatch.org/tigers.htm>, China has an especially shameful record on the importation of animal ingredients, although a similar situation appears to be developing in Japan. Sample (2007) reports that current consumption of bear bile in China was a staggering 4000 Kg in 1998.

Reference.

Sample I. (2007) *Guardian* Tues 12th June 2007.

Next Issue: GM tree culture – what's going on??