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# Rooibos (*Aspalathus linearis*) beyond the farm gate: From herbal tea to potential phytopharmaceutical

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#### Abstract

Aspalathus linearis (Burm.f.) Dahlg. (Fabaceae, Tribe Crotalarieae), an endemic South African fynbos species, is cultivated to produce the well-known herbal tea, rooibos. It is currently sold in more than 37 countries with Germany, the Netherlands, the United Kingdom, Japan and the United States of America representing 86% of the export market in 2010. Its caffeine-free and comparatively low tannin status, combined with its potential health-promoting properties, most notably antioxidant activity, contributes to its popularity. First marketed in 1904 in its fermented (oxidised) form, green rooibos is a new product recently on the market. The utilisation of rooibos has also moved beyond a herbal tea to intermediate value-added products such as extracts for the beverage, food, nutraceutical and cosmetic markets. Its potential as a phytopharmaceutical, shown in recent scientific studies, has not yet been exploited. This review focuses on past and current research aimed at enhancing the value of rooibos herbal tea as a specialised, niche product and expanding its value-adding potential against the background of its traditional use and the current market. The focus falls specifically on aspects such as composition, processing, quality and rooibos as food and potential medicine.

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Keywords: Aspalathus linearis; Herbal tea; Industry; Phytopharmaceutical; Processing; Quality; Rooibos; Value-addition

#### 1. Introduction

The genus *Aspalathus* (Fabaceae, Tribe Crotalarieae) comprises more than 270 species of which most are endemic to the Cape Floristic Region (Dahlgren, 1968). Rooibos tea, produced from *A. linearis* (Burm.f.) Dahlg., had no commercial value at the beginning of the 20th century, but today it is a well-known herbal tea, enjoyed in more than 37 countries. A recent report by the Swiss Business Hub South Africa (Anon, 2007) stated that 'Rooibos appears to be headed towards becoming the second most commonly consumed beverage tea ingredient in the world after ordinary tea (*Camellia sinensis*)'. In South

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Africa it is well established, enjoying popularity amongst an estimated 10.9 million households (data supplied by SA Rooibos Council (SARC), 2011).

Several reviews dealing with rooibos have been published over the years since Cheney deemed it of too insignificant economic importance as a beverage to include it in a review of the biology and economics of the global beverage industry (Cheney, 1947). A different point of view, appreciating its value as a beverage with wide appeal, was later offered (Cheney and Scholtz, 1963). In her review, Morton (1983) campaigned for wider recognition and better distribution of rooibos in the United States of America. It continued to fascinate researchers and recent reviews cover diverse topics such as the development path of rooibos (Rampedi and Olivier, 2008), bioactivity and potential health benefits (Joubert et al., 2008a; Marnewick, 2010; McKay and Blumberg, 2007), and production, processing and quality aspects of rooibos tea and related products (Joubert and Schulz, 2006; Joubert et al., 2008a). The relevance of the

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major rooibos flavonoids in the diet in terms of their bioactivity, bioavailability and potential herb-drug interactions were addressed by Joubert et al. (2009a). The major focus of the present review will be on past and current research aimed at enhancing the value of rooibos herbal tea as a specialised, niche product and expanding its value-adding potential against the background of its traditional use and the current market. Within the context of enhancing the value of rooibos, aspects such as composition, processing, quality, and rooibos as food and potential medicine (phytopharmaceutical) will be discussed.

#### 2. Utilisation of Aspalathus linearis

The first person to realise the commercial potential of rooibos as a herbal tea was Benjamin Ginsberg, a merchant of Clanwilliam, who started marketing it in 1904. He obtained the tea from descendants of the Khoi who crudely processed it during the warm summer months. It was, however, only by 1930 that the agricultural value of rooibos was recognised by a medical practitioner and nature lover, P. Le Fras Nortier of Clanwilliam. His early cultivation experiments, carried out with the help of local farmers, O. Bergh and H. Riordan (Anon, 1985), laid the foundation for the industry. Growing participation of other farmers in rooibos production and increased demand over the years expanded the area under cultivation to 36 000 ha (Pretorius, 2007), with production mainly concen-

trated in the Clanwilliam area (Fig. 1). Since expansion has threatened biodiversity of the Greater Cederberg Biodiversity Corridor, SARC in partnership with CapeNature, formed the Rooibos Biodiversity Initiative. One of its aims was to undertake a joint planning process for expansion to minimise loss of threatened natural habitat (Pretorius, 2007). Following this, a current initiative of SARC is to establish the Right Rooibos Sustainability Standard (http://sarooibos.org.za).

In the early years, different *Aspalathus* species and ecotypes, naturally occurring in the Cederberg mountain area, were used to produce rooibos tea (Dahlgren, 1968, 1988). Today only the so-called red type or Rocklands type, originally from the Pakhuis Pass area, is of commercial importance. The red type is divided into the selected, improved Nortier type (cultivated; Fig. 2), and the Cederberg type (wild-growing). The latter type has broader and coarser leaves than the Nortier type (Morton, 1983). Characteristics associated with the cultivated type are bright green, needle-like leaves on straight, slender branches with relatively short internodes. The leaves should turn redbrown when bruised (Dahlgren, 1968).

Tea is also sometimes made from small quantities of a closely related species, *A. pendula* Dahlg. and several wild types of *A. linearis* (Van Heerden et al., 2003). The grey, black and red-brown types were harvested in the wild, processed and sold to the Rooibos Tea Control Board until 1966, after which the marketing of the grey and black types were discontinued due



Fig. 1. Production areas of Aspalathus linearis in and around the Greater Cederberg Biodiversity Corridor (map supplied by SARC).



Fig. 2. Rooibos plantation in the Clanwilliam area.

to poor quality (Anon, 1967). According to Coetzee et al. (1953) the infusion of the black type was not typically redbrown, nor was the flavour characteristic. Recent assessment of wild types demonstrated morphological (Malgas et al., 2010), genetic (Malgas et al., 2010), chemical (Van Heerden et al., 2003) and ecological (Hawkins et al., 2011) variation.

Wild rooibos re-gained prominence as a source of income in recent years. Small-scale producer organisations in the Cederberg (Wupperthal) and Southern Bokkeveld (Heiveld) supply wild harvested rooibos under organic and fair-trade certification to niche markets abroad (Nel et al., 2007). Wild rooibos comprises ca 2-5% of the annual production of 40 tons rooibos by the Heiveld Co-operative (Patrickson et al., 2008). Wupperthal's production of wild-harvested rooibos is approximately 10 tons (Binns et al., 2007).

# 3. Rooibos tea market

The early years of the industry saw a growing demand due to a shortage of Oriental tea (Camellia sinensis) as a result of the Second World War, followed by market collapse in the years after the war. The Clanwilliam Tea Co-operative Company was subsequently established in 1948 to improve marketing conditions for rooibos tea producers. However, a further market decline made rooibos production uneconomical by 1953/54, which led to the establishment of the Rooibos Tea Control Board (later known as the Rooibos Tea Board), at the request of producers, to stabilise the industry. It started functioning on 6 December 1954 with the objectives, amongst others, to regulate the marketing of rooibos and ensure price stabilisation. Total sales were only 524 tons in 1955. When the Rooibos Tea Board was abolished on 1 October 1993 total sales were more than 4000 tons. The entry of new marketing companies and access to new markets resulted in strong growth in sales during subsequent years. With the abolishment of the Rooibos Tea Board, no organisation dealing with matters of mutual concern existed. This changed in 2005 when SARC was established to coordinate activities relating to generic marketing, research and development, and sustainable natural resource management.

From the modest beginning of Benjamin Ginsberg's marketing effort in 1904 rooibos has become a global product, contributing an estimated R120 million in foreign earnings in 2010 (data supplied by SARC, 2011). Table 1 gives a snapshot of sales (local, export and total) for a 23-year period (1970–

Table 1

Local, export and total sales of rooibos tea (tons) from 1970 to 1992 (data compiled from Anon, 1992).

Year	Local	Export	Total
1970	1277	33	1310
1971	1322	39	1361
1972	1209	49	1258
1973	1088	64	1152
1974	1179	62	1242
1975	1530	86	1616
1976	1440	104	1544
1977	2572	74	2646
1978	3224	140	3363
1979	2186	95	2281
1980	2147	86	2233
1981	2745	140	2885
1982	2086	132	2218
1983	2588	142	2730
1984	2600	177	2777
1985	2378	268	2646
1986	2772	373	3146
1987	2999	316	3315
1988	3123	368	3491
1989	3033	260	3292
1990	3344	432	3776
1991	3292	505	3796
1992	3475	511	3986

1992) preceding the abolishment of the Rooibos Tea Board (Anon, 1992). In 2010, the international market was 5633 tons (data supplied by SARC) compared to 750 tons in 1993 (Anon, 1994). During the past 10 years exports peaked at 5964 tons in 2003 and at 7176 tons in 2007 (Fig. 3). Factors contributing to the decline in export during the past 3 years were global economic uncertainty and a strong Rand (Anon, 2011a).

The major export market through the years remained Germany (previously West Germany), where it was first sold in 1961 as 'Rotbuschtee' or 'Massai-Tee' (Joubert and Schulz, 2006). Exports to Germany as a percentage of the total peaked in 2003 at 76%, thereafter declining to 43% in 2010 (Table 2). During the past 10 years exports to the United States of America (USA) increased more than five-fold from 67 tons to 346 tons. During the same period, exports to the United Kingdom (UK) increased 10-fold from 75 tons to 772 tons. Consumer awareness of rooibos overseas has been boosted, amongst others, by a fictional character, the rooibos tea-drinking Mma Ramotswe, in the 'The No. 1 Ladies' Detective Agency' novels (Goddard, 2005; Jaganath and Crozier, 2010).

The domestic market for rooibos showed a steady increase over the years. In 1984 sales were 2600 tons (Table 1) and rooibos had a 12% share of the domestic tea market (Anon, 2001). By 2010 its market share had more than doubled, representing approximately 23% of the tea market with sales reaching more than 5000 tons with an estimated retail sales value of R429 million (data supplied by SARC).

Organic production of rooibos still lags behind production of conventional rooibos. Exports of organic rooibos reached 828 tons in 2010 (Table 3) with Japan the major market at 222 tons followed by Germany, UK, Netherlands and USA (data from Perishable Products Export Control Board (PPECB), supplied by SARC). Total exports of green rooibos reached 253 tons in 2010 (Table 3), of which 70% went to Germany. The only other country that buys substantial quantities is the Netherlands (16% in 2010). The combined exports of green rooibos to Japan, USA and UK comprised 12% of the total volumes for 2010.

A characteristic of the rooibos industry is the cycles of a shortage in supply, accompanied by high prices, followed by expansion of production and subsequent over-supply, accompanied by low to very low prices. In 2004 producers earned on



Fig. 3. Total export volumes for rooibos during the period 1999–2010 (data from PPECB, supplied by SARC).

average an all-time record of R16.00/kg, but it was followed by a steady decline to R4.50/kg in 2010, a farm gate value last received in 1999 (data supplied by SARC). This negatively affects the economic viability of the crop. Droughts usually contribute to the shortages. In 1984, following overproduction during that production season and the two preceding seasons, the amount of stocks in reserve was ca 10,000 tons (Joubert, 1994) at a stage when total annual sales were less than 3000 tons (Table 1). By 1994, the increased demand could not be met due to a shortage in production (Anon., 1994), yet in 1996 the situation had again changed drastically, i.e. overproduction and low prices. Similar situations have occurred since then, with the most recent record harvests in 2006 (10,000 tons), 2007 (14,000 tons) and 2008 (18,000 tons), respectively, leading to very low prices (Genis, 2008). The production shortage, predicted for 2011, will be the first since 2003 (Swartz, 2011).

### 4. Geographical indication (GI)

Opportunistic trade marking of the term 'rooibos' in 1994 in the USA, which prevented the free use of this generic term in marketing and the subsequent legal battle to reclaim the name for South Africa, has given impetus to the development of a Geographic Indication (GI) for rooibos (Gerz and Bienabe, 2006). It is mainly seen as a defensive measure against possible cultivation outside South Africa and to protect intellectual property rights. A GI is a label that is reserved for products which acquire their characteristic and defining qualities as a result of their geographical location, e.g. Champagne, Tequila or Parma Ham (Grazioli, 2002). In this way producers can distinguish their product based on its specific origin-related characteristics.

The rigorous process of GI certification means that strict quality criteria, linked to its geographic cultivation, must be implemented. Aspalathin, as an unique rooibos phenolic constituent, is also under consideration as a defining parameter. It is believed that these quality criteria will have a positive effect on quality control and marketing (Anon, 2010). Results of an ARC project on the development of the sensory wheel and objective quality criteria (Sections 8.1 and 8.2), undertaken for SARC, will be critical for the certification and required quality control processes.

#### 5. Value addition

The rooibos industry, characterised by bulk sales as opposed to the sale of retail packaged products to international markets, would greatly benefit from value-addition (Anon, 2010). Based on the number of patents filed (Kaplan, 2009), protection of intellectual property for new applications and/or processes relating to rooibos were considered worth-while. Japan has been especially active in this regard.

The first value-addition was done by A. Theron who brought an extensive cosmetic care range containing rooibos extract to the market (Joubert and Schulz, 2006). Her range of products is distributed to 34 countries (Anon, 2010). Scientific studies in

Table 2 Major export countries with % of export to each from 2001 to 2010 (data from PPECB, supplied by SARC).

•	*		,			• /				
Country	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010
Germany	57.50	64.91	75.88	74.01	63.25	56.90	54.83	50.76	48.17	42.82
Netherlands	14.89	15.75	7.63	9.47	13.48	13.54	10.88	10.95	15.25	17.32
United Kingdom	1.85	3.05	2.84	2.74	5.75	7.22	7.35	11.37	12.56	13.70
Japan	14.94	7.78	7.95	5.21	5.12	5.30	5.76	3.78	6.07	6.34
United States of America	1.72	2.07	2.06	5.01	4.07	6.56	5.34	4.96	4.45	6.14
Other	9.10	6.43	3.63	3.56	8.33	10.47	15.85	18.19	13.50	13.69

the public domain dealing with the topical/cosmetic application of rooibos extract are, however, very limited. The role of aspalathin, the major rooibos flavonoid and antioxidant, has not yet been clarified. Poor penetration of the skin by aspalathin has been demonstrated by Huang et al. (2008a). An anti-wrinkle effect was demonstrated for a formulation containing rooibos and tea (*Camellia sinensis*) (Chuarienthong et al., 2010). Since rooibos extract was not tested alone it is not possible to make conclusions about its efficacy as an anti-wrinkle agent. According to Glynn (2010) the topical application of an herbal mixture, containing green rooibos extract, alleviates male pattern baldness. A recent US patent application deals with the protective effect of rooibos extract against hair colour loss (Joppe et al., 2009).

Rooibos extracts, produced locally and abroad, are considered intermediate value-added products in the value chain. Apart from cosmetics, final product applications include functional foods and beverages, as well as nutraceuticals. Rooibos is also finding application in pet food and pet skin care products (Anon, 2010).

One of the earliest applications of rooibos extract and its volatile fraction was as a flavour ingredient of yogurt by the then Van Riebeeck Dairies when the rooibos instant tea powder was first developed (Joubert, 1984). At that stage the use of rooibos as functional ingredient was not yet realised. Neither the instant rooibos tea nor the yogurt reached the market. It was only in 2000 that the manufacture of rooibos powdered extract by a South African company became a reality, when changing consumer attitudes towards natural products made it a viable value-addition option.

The types of rooibos extract and the raw material from which it is produced depends on the final application. A broad distinction can be made between extracts from fermented and green rooibos. Fermented rooibos provides the bulk of the material for production of extracts, used predominantly in

Table 3 Exports of organic and green rooibos (tons) from 2003 to 2010 (data from PPECB, supplied by SARC).

, F F	J ··· - ) ·	
Year	Organic	Green
2003	648	83
2004	382	60
2005	624	64
2006	386	112
2007	756	74
2008	840	87
2009	791	145
2010	828	253

beverages and functional foods. Its aroma extract is used to flavour beverages (Anon, 2010).

Some of the food and beverage products currently on the market that contain rooibos extract are yogurt, drinking yogurt, ready-to-drink iced tea, jam and 'instant cappuccino'. Labelling of the rooibos-containing yogurt, carrying the Cancer Association of South Africa Smart Choice emblem, makes extensive use of potential functional properties of rooibos. The drinking yogurt also refers to the 'active ingredient' of rooibos, but no specific 'active ingredient' is mentioned. It must be assumed that the 'active ingredient' is the rooibos solids as no single compound in rooibos is solely responsible for its antioxidant and anti-cancer properties (Joubert et al., 2009a; Snijman et al., 2007).

In many instances, rooibos is only one of several herbal and fruit ingredients in beverages (Anon, 2009a, b). Functional drinks launched in USA and Italy in 2009 contain, amongst others, rooibos and pomegranate. Another drink, launched in the same year in Japan by a major soft drink company contains, apart from rooibos as main ingredient, also green tea, Japanese citrus fruit peel, kumazasa (bamboo), pure tea, fennel, turmeric and hawthorn extract.

Green rooibos, first produced as part of an ARC research project during the 1990s to achieve higher antioxidant levels, is used as a tea and for preparation of extracts for the food, cosmetic and functional beverage markets. The higher levels of flavonoids (antioxidants) in green rooibos, combined with its caffeine-free status contribute to its popularity in tea blends (Cosgrove, 2010) and cosmetic products (Otto et al., 2003; Tiedtke and Marks, 2002). Aspalathin-enriched extracts can be prepared from green rooibos as this compound is present in high levels (Manley et al., 2006; Schulz et al., 2003). The level of enrichment greatly depends on extraction conditions and level of purification. According to a patented process, enrichment is achieved with organic solvents (Grüner-Richter et al., 2008). The tan to greenish coloured extract was initially developed for the cosmetic market (B. Weinreich, Raps & Co, Germany; pers. comm.). A Japanese company also produces an orange coloured aspalathin-enriched extract, specified to contain a minimum of 20% aspalathin. This extract is destined for the supplement market (H. Hata, Tama Biochemical Co., Japan; pers. comm.).

Pharmaceutical product applications of rooibos have not yet been exploited, but according to the report by Kaiser Associates (Anon, 2010) opportunities are likely to emerge as a result of the trend towards phytopharmaceuticals. Rooibos extracts, usually combined with other ingredients, are available in pill form, but these products fall in the category of dietary supplements. Recent research has underscored the potential of aspalathin and

Table 4
Contents of major phenolic compounds in green and fermented Aspalathus linearis plant material <sup>a</sup> (means±SD; g/100 g dry matter).

Structures	Compounds	Green (n=3)	Fermented (n=3)
HO OH R1 R2 OH O	Dihydrochalcones aspalathin ( $R_1$ =OH; $R_2$ = <i>C</i> -glucosyl) nothofagin ( $R_1$ = <i>C</i> -glucosyl; $R_2$ =OH)	$2.559 \pm 0.699$ $0.251 \pm 0.230$	$0.421 \pm 0.017$ $0.040 \pm 0.022$
$R_2$ $OH$ $R_4$ $R_3$ $OH$ $OH$ $OH$	Flavones orientin ( $R_1=C$ -glucosyl; $R_2$ , $R_4=OH$ ; $R_3=H$ ) iso-orientin ( $R_1=H$ ; $R_2$ , $R_4=OH$ ; $R_3=C$ -glucosyl) vitexin ( $R_1=C$ -glucosyl; $R_2=OH$ ; $R_3$ , $R_4=H$ ) isovitexin ( $R_1$ , $R_4=H$ ; $R_2=OH$ ; $R_3=C$ -glucosyl) luteolin ( $R_1$ , $R_3=H$ ; $R_2$ , $R_4=OH$ ) luteolin-7- $O$ - $\beta$ -D-glucoside ( $R_1$ , $R_3=H$ ; $R_2=O$ -glucosyl; $R_4=OH$ ) chrysoeriol ( $R_1$ , $R_3=H$ ; $R_2=OH$ ; $R_4=OCH_3$ )	$\begin{array}{c} 0.263 \pm 0.087 \\ 0.450 \pm 0.163 \\ 0.042 \pm 0.020 \\ 0.049 \pm 0.029 \\ 0.007 \pm 0.006 \\ 0.015 \pm 0.008 \\ 0.003 \pm 0.001 \end{array}$	$0.202 \pm 0.026$ $0.329 \pm 0.049$ $0.035 \pm 0.009$ $0.035 \pm 0.012$ $0.010 \pm 0.005$ $0.015 \pm 0.008$ $0.007 \pm 0.002$
$HO$ $OH$ $OH$ $HO$ $R_1$ $R_2$ $O$ $R_3$	Flavonols quercetin ( $R_1$ =H; $R_2$ , $R_3$ =OH) hyperoside ( $R_1$ =H; $R_2$ =OH; $R_3$ =O-galactosyl) rutin ( $R_1$ =H; $R_2$ =OH; $R_3$ =O- rutinosyl)	$\begin{array}{c} 0.001 \pm 0.001 \\ 0.021 \pm 0.012 \\ 0.245 \pm 0.141 \end{array}$	$0.010 \pm 0.001$ $0.016 \pm 0.015$ $0.173 \pm 0.016$

<sup>a</sup> Fresh plant material from 3 individual plants was divided in two to produce green (dried whole, 40 °C/12 h) and fermented (shredded; fermented, 38 °C/12 h; dried, 40 °C/12 h) plant material.

selected rooibos extracts such as an aspalathin-enriched green rooibos extract as anti-diabetic agents (Joubert et al., 2010a,b; Kawano et al., 2009; Mose Larsen et al., 2008). A patent application for the use of aspalathin in this context was filed in Japan (K. Yagasaki, TUAT, Japan; pers. comm.), while a PCT application was filed for the use of rooibos extract as an antidiabetic agent (Mose Larsen et al., 2008). It is claimed that rooibos extract and a heterodimer containing aspalathin, isolated from rooibos, could be used as a medicament for the treatment of neurological and psychiatric disorders of the central nervous system (Bruno and Dimpfel, 2009). Other opportunities may lie with topical skin products. Two studies concerning inhibition of COX-2 in mouse skin (Na et al., 2004) and inhibition of mouse skin tumour promotion (Marnewick et al., 2005), support the role of the topical application of rooibos extract in preventing skin cancer.

## 6. Composition

Rooibos is prized as a caffeine-free herbal tea, although traces of the alkaloid sparteine have been reported (Van Wyk and Verdoorn, 1989). Despite being well-known as a low tannin tea, ca 50% of the hot water soluble solids is tannin-like substances (Joubert et al., 2008a). When compared to black tea (*Camellia sinensis*), rooibos contains less tannins (Blommaert and Steenkamp, 1978). Very little information is available on the structure of the tannins from rooibos tea, but the dimer, procyanindin B3, the trimer, bisfisetinidol- $(4\beta, 6:4\beta, 8)$ -catechin, and a pentamer have been identified (Ferreira et al., 1995).

Rooibos contains two unique phenolic compounds, namely aspalathin (Koeppen and Roux, 1965), a dihydrochalcone C-glucoside, and aspalalinin (Shimamura et al., 2006), a cyclic dihydrochalcone. Nothofagin, previously only identified in the heartwood of Nothofagus fusca (Hillis and Inoue, 1967) and the bark of a Chinese medicinal plant Schoepfia chinensis (Huang et al., 2008b), is a rare dihydrochalcone C-glucoside present in rooibos. Other major phenolic compounds present in rooibos include flavones (orientin, isoorientin, vitexin, isovitexin, luteolin, chrysoeriol), flavanones (dihydro-orientin, dihydro-isoorientin, hemiphlorin) and flavonols (quercetin, hyperoside, isoquercitrin, rutin) (Ferreira et al., 1995; Koeppen et al., 1962; Marais et al., 2000; Rabe et al., 1994; Shimamura et al., 2006). Phenolic acids, lignans, flavone diglycosides, (+)-catechin, a phenylpyruvic acid glycoside, the flavonol quercetin-3-O-robinobioside and the coumarins, esculetin and esculin, have also been identified (Beltrán-Debón et al., 2011; Breiter et al., 2011; Krafczyk and Glomb, 2008; Marais et al., 1996; Shimamura et al., 2006).

Fermentation of rooibos plant material to produce the traditional herbal tea (oxidised form) causes substantial quantitative changes in its phenolic composition (Table 4, unpublished data). One of the major changes is oxidation of aspalathin via its flavanone analogues to isoorientin and orientin (Koeppen and Roux, 1965; Marais et al., 2000). The oxidation mechanism was recently further elucidated by Krafczyk and



Fig. 4. Mechanism of aspalathin oxidation (compiled from Krafczyk and Glomb (2008) and Krafczyk et al. (2009)).

Glomb (2008) (Fig. 4). Dihydro-isoorientin ((R)- and (S)eriodictyol-6-C-glucoside), preferentially formed from aspalathin, oxidises to isoorientin. However, dihydro-orientin ((R)and (S)-eriodictyol-8-C-glucoside) does not oxidise directly to orientin as is the case with dihydro-isoorientin and its flavone, isoorientin. Orientin forms irreversibly from isoorientin, which undergoes opening of its vinyl ester structure to form a chalcone intermediate, which is then converted to orientin. Furthermore, an aspalathin dimer, partly responsible for browning as a result of aspalathin oxidation, has also been identified (Krafczyk et al., 2009).

The potential health benefits of rooibos tea have been linked to its phenolic content. However, insufficient data are available on the dietary exposure to rooibos phenolic compounds (Joubert et al., 2009a). The calculation of rooibos phenolic intake in populations will allow a study of their association with health and disease. For this purpose, it is essential to have detailed and comprehensive information on the nature and quantities of phenolic compounds in rooibos tea. Increasing interest in dietary phenolic compounds and their effect on human health has led to the development of several databases on phenolic compound contents in foods, such as the USDA Databases on Flavonoids, Phenol-Explorer, and EuroFIR-BASIS (Grv et al., 2007; Neveu et al., 2010; Pérez-Jiménez et al., 2010). None of these databases include composition data for rooibos. Current HPLC methods employed for the analysis of rooibos infusions and extracts are only suitable for the quantification of some of the major flavonoids due to the co-elution of various phenolic constituents. Fingerprinting for authentication/QC and accurate quantification are therefore not possible. Recently, a new HPLC method has been developed for the simultaneous quantification of 16 rooibos phenolic compounds (Beelders et al., 2010a). The method is currently being used by the ARC to analyse a large number of infusions prepared from fermented rooibos production batches obtained from the 2009, 2010 and 2011 harvest seasons, to give a snapshot of phenolic variation in rooibos tea infusions as enjoyed by the consumer. This data would be suitable for inclusion in food composition databases. In addition, an even more comprehensive investigation of rooibos phenolic compounds using 2-dimensional liquid chromatographic techniques is underway (Beelders et al., 2010b).

Very little information on the mineral content of rooibos infusions is available. Touyz and Smit (1982) showed that an infusion of fermented rooibos contains  $1.29 \,\mu$ g/mL fluoride.

Interestingly, Joubert et al. (2008a) showed a relatively high sodium content of  $43.33 \,\mu$ g/mL for the infusion. The toxic mineral, aluminium, is present in much lower quantities in fermented and green rooibos infusions than in *Camellia sinensis* infusions (Malik et al., 2008). The contents of mineral nutrients are also fairly low (Joubert et al., 2008a; Malik et al., 2008).

# 7. Processing

# 7.1. Tea

Traditional or conventional rooibos is 'fermented' to develop its characteristic red-brown leaf and infusion colour, and pleasant, slightly sweet flavour. Although modernised, the processing steps employed today are still based on the traditional method of chopping the shoots into small pieces, bruising with wooden hammers to initiate browning, 'sweating' (fermentation) and drying for flavour development (Anon, 1985). Steam pasteurisation has been introduced in the 1980s to ensure good microbial quality after *Salmonella* contamination was found.

A study carried out on processing showed that quality and product consistency could be improved by carrying out fermentation and drying under controlled conditions (Joubert, 1994, 1998; Joubert and De Villiers, 1997; Joubert and Müller, 1997). A factory-based process is, however, not feasible due to the economics of scale and the need for artificial heat generation during drying as opposed to natural heat supplied by sunlight. Observations made by producers and technical personnel of the then Rooibos Tea Board suggest that factors such as the presence of young growth, the age of the bush and the cultivation area could affect the tea quality (Joubert, 1994).

Chemical changes during fermentation are responsible for the development of the red-brown colour (Section 5). Recent papers by Krafczyk and Glomb (2008) and Krafczyk et al. (2009) provide greater understanding of the conversion of aspalathin, previously studied by Koeppen and Roux (1965) and Marais et al. (2000). Aspalathin is not only converted to flavanones as intermediate products, flavones and a dimer, but these constituents are minor products among uncharacterised brown material. Apart from the substantial losses in nothofagin (Joubert, 1996) no other chemical changes during fermentation have been studied yet.

When processing green rooibos, the unfermented product, the greatest challenge is to prevent or to minimise oxidative changes so that the green colour and aspalathin are retained. This could be achieved by inactivation of enzymes, for example, by subjecting the tea to a rapid steaming process. Alternatively, low temperatures and water activity or exclusion of oxygen during drying can be used to slow down the rate of the chemical reactions. A patented vacuum-drying process (De Beer and Joubert, 2002) produced a good quality product with a long shelf-life. Samples stored in a dry, dark place were still green after 2 years (unpublished data). Since vacuum-drying is carried out batch-wise and is expensive, its use did not extend past short-term pilot production of green rooibos. One of the techniques, employed by industry, is to spread the tea in a thin layer in the sun to facilitate rapid drying. Under these conditions, production of good quality green rooibos remains a challenge. If not properly dried, slow browning of the tea will take place during storage and transport to overseas markets, resulting in a product with the appearance of a poorly fermented rooibos, yet without its sought-after characteristic slightly sweet flavour.

# 7.2. Extracts

Waste material in the form of coarse stems and uncut leaves that were sieved out and of no commercial value served as the stimulus of a study carried out in the early 1980s that led to the development of an instant rooibos from the fermented plant material (Joubert, 1984). The idea was also to provide the consumer with rooibos in a more convenient form (Joubert, 1988a) as brewing of a strong cup of tea at that stage was a timeconsuming process. Low recovery of soluble solids from the waste (8.9% versus 20.4% for the sieved fraction < 10 mesh) and overproduction (see Section 3) soon shifted the focus to the tea fraction used for commercial purposes (Joubert, 1984, 1988a). The concept of a soluble rooibos product only found commercial application in 2000 in South Africa with the production of powdered extracts for the beverage, food and dietary supplement markets (Section 5).

The effect of factors such as extraction temperature and time, tea-to-water ratio and extraction process (stirred batch-wise and fixed-bed with flow through) on the recovery of soluble solids and their phenolic composition were investigated (Joubert, 1988a, 1990a,b; Joubert and Hansmann, 1990). Enzyme treatment of the plant material was shown to increase the yield of soluble solids (Pengilly et al., 2008). Normally a muddy brown precipitate forms upon cooling of the extract after concentration. This precipitate was found to impair clarity of the reconstituted extract at 'cup of tea' strength. Its removal improved the clarity, but decreased the phenolic content of the final product (Joubert, 1990c). Enzyme treatment of rooibos before extraction could improve the solubility and clarity of reconstituted rooibos extract (unpublished data). Powdered rooibos extract is not yet 'instant' and agglomeration is required to achieve the characteristics of an instant product, i.e. no lumping when added to hot water and quick dispersal with minimum agitation in the cup (Joubert, 1988b).

Information on the technological processes involved in green tea extract production is not readily available, except when the process is patented. Section 5 provides some discussion on the types of extracts available from green rooibos.

## 8. Tea quality aspects

The current South African regulations relating to quality standards for rooibos deal with moisture content, pesticide residues, microbial contamination and the percentage of white stems allowed (Anon, 2002). Taste, aroma and colour are dealt with in vague, meaningless terms, i.e. rooibos should 'have the clean, characteristic taste and aroma and clear, distinctive colour of rooibos', without definitions or reference standards for the terms 'characteristic' and 'distinctive'. Another description used by industry is 'typical'. This means that manufacturers have the freedom to set their own quality standards in terms of colour, flavour and mouth feel of rooibos infusions. Furthermore, the quality classifications 'Super' and 'Choice' only have meaning within the context of a specific retail brand.

In the past the 'caffeine-free' status and 'low tannin content' of rooibos formed the basis of its 'good-for-you' message to consumers. Presently, the antioxidant activity of rooibos, in addition to its pleasant flavour, contributes to its popularity on the international markets. However, rooibos is competing in a market cluttered with products 'rich in antioxidants' or a market characterised by re-positioning of existing products such as the South American herbal tea, yerba mate, with a high caffeine content. A quick survey of scientific literature during the past 5 years will show that the number of papers dealing with plant extracts, whether from fruits or herbs, and their antioxidant activity have increased dramatically. A number of studies included rooibos for comparative purposes and several concluded that their extracts have higher antioxidant activity than rooibos (e.g. Ivanova et al., 2005; Piccinelli et al., 2004; Yoo et al., 2009). In the face of such competition, rooibos tea quality should therefore take on a new meaning (Sections 8.1 and 8.2).

## 8.1. Sensory quality

For descriptors of rooibos sensory properties to have any value, 'characteristic', 'typical' and 'distinctive' must have the same meaning for all role players in the rooibos industry, including the global industry. For this reason a flavour and mouthfeel wheel, which incorporates both positive and negative sensory attributes, totalling 17 descriptors, was developed for rooibos (Anon, 2011b). A large number of samples was analysed to derive the final, most frequently occurring, descriptors. Fine-tuning of the wheel by including data from a second harvest season is in progress. To aid interpretation of the descriptors has also been developed (Koch, 2011). A sensory lexicon is a set of words that describe the sensory attributes of a product, along with definitions and/or reference standards for clarification (Drake and Civille, 2002).

The sensory wheel will provide better understanding and appreciation for rooibos sensory characteristics. This will create opportunities for niche products with specific flavours such as estate teas, or teas originating from a specific area as climatic and soil conditions could contribute to different dominant flavour nuances, e.g. caramel or floral.

#### 8.2. Other quality parameters

The rooibos industry is increasingly interested in using polyphenol content and antioxidant activity for marketing purposes, as well as for quality control of extracts for the nutraceutical and cosmeceutical markets. Large variation in the antioxidant activity of different production batches of rooibos could, however, erode customer confidence in the product, especially customers who are interested in producing valueadded products such as rooibos extract powders that should meet a predetermined antioxidant activity level. By grading rooibos based on composition and/or antioxidant activity levels, marketers would be able to guarantee minimum levels and product consistency. Producers would be able to blend different batches to reach a predetermined antioxidant level. Against this background, three potential objective quality parameters, i.e. water-soluble solids content, total antioxidant capacity (TAC) and total polyphenol content (TPC) are currently under investigation by the ARC.

Two of the prominent South African extract producers use TPC and TAC as indicators of quality. A major extract manufacturer also produces an extract standardised on orientin and isoorientin content (>0.5% total), while an extract of fermented rooibos, standardised on aspalathin content, is under development (D. Malherbe, Afriplex, Paarl, South Africa; pers. comm.).

Many researchers have shown that for a specific type of product various antioxidant assays correlate well with TPC (e.g. Prior et al., 2005). Very good correlations between TPC and ABTS<sup>•+</sup> scavenging activity were also obtained for unfermented rooibos water extracts (r=0.99; Joubert et al., 2008b). Interestingly, aspalathin content of unfermented rooibos water extracts was also highly correlated with ABTS<sup>•+</sup> scavenging activity (r=0.96; Joubert et al., 2008b).



Fig. 5. Variation of aspalathin (A) and orientin (B) content (g/100 g dry matter) in leaves of 21 plants harvested at the same time from a single plantation (aspalathin  $\blacklozenge$  data from Joubert et al., 2008c; orientin  $\blacklozenge$  unpublished data).

Care should be taken when interpreting antioxidant values. It is especially important to be careful of directly comparing antioxidant results from different sources as differences in assay protocol between studies can have a large influence on the values obtained. A myriad assays are available for determining the antioxidant activity of foods and beverages. Prior et al. (2005) reviewed the assays most commonly used for analysis of foods and dietary supplements and suggested the ORAC (Oxygen Radical Absorbance Capacity) (Ou et al., 2001), ABTS<sup>++</sup> scavenging (Re et al., 1999) and Folin-Ciocalteau (Singleton and Rossi, 1965) assays based on their advantages and disadvantages. Generally the ORAC assay is favoured by the American nutraceutical industry to 'quantify' the total antioxidant capacity of extracts (Crawford, 2011).

### 8.3. Plant material

The composition of the plants has a direct effect on the quality of processed fermented tea and commercial extracts. Rooibos plants are cultivated from seedlings, causing large variation in phenolic composition between plants. As an example the aspalathin and orientin content of rooibos leaves harvested at the same time from 21 individual plants in the same plantation is shown in Fig. 5A (data adapted from Joubert et al., 2008c) and 5B (unpublished data), respectively. The aspalathin and orientin content ranged from 6.0 to 11.2 g/100 g dry matter and 0.5 to 1.1 g/100 g dry matter, respectively. Harvest date is another factor that could influence phenolic composition of rooibos plants. The aspalathin content of plant material harvested from the same plants ranged from 2.33 to 3.76 g/100 g dry matter over a one year period (unpublished data).

The phenolic composition of wild rooibos showed qualitative and quantitative variation between populations and types (Van Heerden et al., 2003). Some populations even lacked aspalathin, the major flavonoid in the red type or Rocklands type of *A. linearis* used for commercial production. One of the wild types, lacking aspalathin, contained rutin as major flavonoid (Joubert and Schulz, 2006). If the presence of aspalathin in the plant material (green and fermented) becomes part of the quality parameters of a rooibos GI (Section 4) then some wild rooibos types may be excluded from the 'rooibos' label in future.

# 8.4. Commercial extracts

Recently, changes in quality of extracts during the manufacturing process were investigated in terms of aspalathin, orientin, isoorientin and total polyphenol contents (Joubert et al., 2009b; Viljoen, 2008). No major losses of aspalathin, orientin or isoorientin were observed during microfiltration, reverse osmosis and concentration of fermented rooibos extract. The total polyphenol content, on the other hand, seemed to increase during these processes (Viljoen, 2008) indicating that compounds other than polyphenols are removed during the production process. Spray-drying of the concentrate did not affect its phenolic content.

#### 8.5. Ready-to-drink iced teas

Ready-to-drink rooibos iced tea is a popular variant in South Africa; however, its rooibos content is not regulated. A recent study (Joubert et al., 2009b) found that some brands contained no aspalathin or its oxidation products, orientin and isoorientin, suggesting that no rooibos extract was used in their manufacture. The maximum aspalathin content found in 41 samples, representing eight brands, was 0.69 mg/100 mL. The average aspalathin content of the samples was 0.28 mg/100 mL, which was much lower than that expected in rooibos extract reconstituted to 'cup of tea' strength (0.3–0.9 mg/100 mL; n=20; unpublished data).

In an attempt to understand the low levels of rooibos flavonoids in commercial iced teas, a study was conducted to assess their stability in fermented (Joubert et al., 2009b) and green (Joubert et al., 2010c) rooibos iced tea formulations



Fig. 6. Anti-ageing effect of rooibos on the human skin. Cartoon originally published in Landbouweekblad of 6 March 1992 (reproduced with permission from Landbouweekblad).

Table 5

S	ummarv	of it	ı vitro	and in	vivo studies	s on the	bioactivity	of rooil	oos infus	sions or ex	tracts pub	olished	after th	ne review	of Joubert	et al., 20	.)08a.
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Bioactivity	Infusion/ extract	Model	Outcome	Reference
Regeneration of	Fermented a aq. extract	CCl <sub>4</sub> -induced liver damage in rats	Reduced activity of ALT and AST;	Uličná et al.,
liver damage			reduced total bilirubin;	2008
Anti-inflammatory	Unfermented ag extract	Wistar rat DSS-induced colitis	Increased SOD levels:	Baba et al
activity	emermented aq. enalaet		decreased 8-hydroxy-2'-	2009
			deoxyguanosine levels	
Chemoprotection	Unfermented and	Oral administration to Fischer rats;	Fermented tea decreased lipid	Marnewick
	termented aq. infusions	$FB_1$ and DEN, respectively	unfermented tea reduced total	et al., 2009
			number if foci and relative	
			amount of large foci; both teas	
			levels: fermentation reduced	
			chemoprotective activity	
Tumouricidal	Fermented <sup>a</sup> EtOH extract	Murine neuroblastoma cell line	Weak activity based on $LC_{50}$ >	Mazzio and
activity		derived from a spontaneous malignant tumour	5 mg/mL	Soliman, 2009
PPAR $\gamma$ ligand	Fermented a DMSO extract	Polar Screen PPAR $\gamma$	Moderate ligand-binding activity	Mueller and
binding activity		ligand-binding		Jungbauer,
		competitive assay	Strong antagonist of coactivator	2009 Mueller and
		coactivator assay	recruitment to PPAR $\gamma$	Jungbauer,
		2		2009
		Transactivation activity using a	No transactivation activity	Mueller and
		chimeric GAL4- PPARy assay		2009
Inhibition of	Fermented and	Inhibition of substrate binding assay	Competitive inhibition; unfermented	Perold, 2009
adrenal	unfermented aq. and	to CYP17 and CYP21 in adrenal	MeOH extract more	
steroidogenic P450 enzymes	MeOH extracts	microsomes Inhibition of substrate metabolism in	effective than termented MeOH extract Stronger inhibition of CYP21:	Perold 2009
1 100 011251100		COS1 cells expressing baboon	fermented weaker than	101010, 2009
		CYP17 or CYP21	unfermented	
Anti-carcinogenic	Unfermented and fermented EtOH extracts	Two-stage skin carcinogenesis	Reduced incidence and	Petrova, 2009
photoprotective	Termented Etorr extracts	promotor and initiator, respectively	erythema, peeling, oedema and	
effect		· · · · ·	hyperplasia; depletion of	
			catalase and SOD prevented;	
			DNA damage; reduced lipid	
			peroxidation; reduced	
Ori lation stores	F		induction of COX-2 and ODC	W
reduction	Fermented aq. infusion	Single oral dose to healthy humans	capacity and GSH/GSSH	Wanjiku, 2009
Toddettoll			erythrocyte ratio	
Cytoprotective	Fermented <sup>a</sup> 70% aq.	Chinese hamster lung fibroblast	Reduced H <sub>2</sub> O <sub>2</sub> induced cell	Yoo et al.,
activity	MeOH extract	V/9-4 cells	membrane damage; reduced intracellular ROS generation	2009
Anti-wrinkle	Commercial extract	Topical application to humans	Reduced wrinkles; no effect	Chuarienthong
activity	blend with Camellia	(female)	on skin smoothness,	et al., 2010
Inhibition of	sinensis extract	Single and dose containing sugar	scaliness and roughness Reduced plasma glucose	Francisco
post-prandial	i ennemed aq. musion	with a high fat meal to	insulin, total cholesterol, LDL	2010
oxidative stress		healthy humans	cholesterol, triglycerides,	
			hsCRP, CD and TBARS levels;	
			capacity and total glutathione	
			level	
Promotion of hair	Blend of botanical	Topical application to	Increased hair density,	Glynn, 2010
growin	unfermented rooibos	with androgenetic alopecia	hair growth rate	
		(male pattern baldness)	g	
Anti-inflammatory	Fermented <sup>a</sup> aq. infusion	Whole blood culture assay;	Increased secretion of IL-6, IL-10,	Hendricks and
activity		unstimulated, endotoxin	and IFN $\gamma$ in unstimulated	Pool, 2010

(continued on next page)

#### Table 5 (continued)

Bioactivity	Infusion/ extract	Model	Outcome	Reference
		stimulated and PHA stimulated	cells; increased IL-6, decreased IL-10 and no change for TNFy secretion in stimulated cells	
Hypoglycaemic activity	Fermented aq. extracts and aspalathin-enriched unfermented extracts	STZ-induced hyperglycaemic Wistar rats	Lowered blood glucose levels	Joubert et al., 2010b
	Fermented aq. extract	Diet-induced type II diabetic vervet monkeys	Lowered blood glucose levels	Joubert et al., 2010b
Anti-inflammatory activity	Fermented <sup>a</sup> DMSO extract	LPS-stimulated macrophages	Reduce secretion of IL-6 and IL-10; increased expression of COX-2; no effect on TNFα secretion and iNOS expression	Mueller et al., 2010
Angiotensin- converting enzyme inhibition	Fermented <sup>a</sup> aq. infusion	Single oral dose to healthy humans	Inhibited ACE activity for the ACE II genotype; no effect on nitric oxide concentration	Persson et al., 2010
Rehydration	Fermented <sup>a</sup> aq. infusion	Oral administration to humans after 2 h wrestling practice	Rehydration due to rooibos comparable with water control	Utter et al., 2010
Oxidative stress reduction and lipid profile modulation	Fermented and unfermented aq. extracts in ready-to-drink beverage	Single oral dose to healthy humans	Increased plasma antioxidant capacity; no change in lipid profile or uric acid level	Villaño et al., 2010
Anti-hyperlipidaemic activity	Fermented <sup>a</sup> aq. extract	Hyperlipidaemic male LDLr–/– mice	Reduced serum cholesterol, triglyceride and free fatty acid levels; prevention of dietary-induced hepatic steatosis	Beltran-Debon et al., 2011
Anti-adipogenesis activity	Fermented <sup>a</sup> aq. extract	3T3-L1 mouse adipocytes	Dose-dependent inhibition of triglyceride accumulation	Beltrán-Debón et al., 2011
Oxidative stress reduction and lipid profile modulation	Fermented aq. infusion	Oral administration to humans with cardiovascular risk factors over a 6 week period	Decreased CD, TBARS, LDL cholesterol and triacylglycerols levels; increased GSH level, GSH/GSSG ratio and HDL cholesterol	Marnewick et al., 2011

aq., aqueous; ACE, angiotensin-converting enzyme; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CD, conjugated dienes; COX, cyclooxygenase; DEN, diethylnitrosamine; DMBA, 7,12-dimethylbenz[a]anthracene; DMSO, dimethylsulphoxide; DSS, dextran sodium sulphate; EtOH, ethanol; FB<sub>1</sub>, fumonisin B<sub>1</sub>; GSH, reduced glutathione; GSSG; oxidised glutathione; HDL, high-density lipoprotein; hsCRP, high sensitive C-reactive proteins; IFN, interferon; IL, interleukin; iNOS, inducible nitric oxide synthase; LC<sub>50</sub>, concentration needed to kill 50% of cells; LDL, low-density lipoprotein; LPS, lipopolysaccharide; MeOH, methanol; ODC, ornithine decarboxylase; PHA, phytohemagglutinin; PPAR, peroxisome proliferators-activated receptor; SOD, superoxide dismutase; STZ, streptozotocin; TBARS, thiobarbituric acid reactive substances; TNF, tumour necrosis factor; UVB, ultraviolet B light.

<sup>a</sup> Fermentation state of plant material used is not specified, but fermented tea is assumed as this is the form most commonly sold.

during heat treatment. Pasteurisation or sterilisation is the most likely processing step during the manufacture of rooibos iced teas that could decrease their flavonoid content. Pasteurisation caused no loss of rooibos flavonoids in fermented rooibos iced tea, but sterilisation markedly decreased the aspalathin and isoorientin contents (Joubert et al., 2009b). The presence of citric and ascorbic acids partially protected against the loss of aspalathin and isoorientin. Orientin showed different trends due to its conversion from isoorientin. The aspalathin content of iced tea could be greatly increased by using an aspalathinenriched extract produced from green rooibos (Joubert et al., 2010c). Trends for heat stability of the flavonoids in this type of rooibos iced tea were similar to that of fermented rooibos iced tea. Combining the aspalathin-enriched green rooibos extract with ascorbic acid in a nano-micelle carrier system increased the heat stability of the resulting iced tea formulation with only a minor reduction in aspalathin loss after addition of citric acid.

Results from these two studies show that rooibos iced tea could make a valuable contribution to aspalathin intake if care is taken during formulation and processing. The bioavailability of aspalathin when present in a ready-to-drink rooibos iced tea was recently demonstrated (Section 9).

Storage of iced tea can also contribute to low levels of aspalathin. Its stability during storage was shown to depend on product formulation and type of extract (Viljoen, 2008). pH was shown to be a crucial factor in its stability (De Beer et al., 2011). Milk-based products with a relatively high pH would not retain aspalathin.

### 9. Food or medicine conundrum

The discovery by A. Theron in 1968 that rooibos helped to soothe her allergic baby's colic started off rooibos tea's reputation as a 'healthy drink' in modern times (Joubert and Schulz, 2006). Her anecdotal findings, summarised in a letter to the Rooibos Tea Control Board on 18 May 1968, was met with scepticism from scientists who were approach for comment. More anecdotal evidence became evident after newspaper reports and radio interviews and today the therapeutic value of rooibos for colic babies is well-known.

In the 1985 Annual Report of the Rooibos Tea Board (Anon, 1985) it was noted that: 'Rooibos Tea is used overseas mainly for its medicinal value and has thus become popular as a medicine rather than a tea. This is to a certain extent the main reason why overseas marketing potential of Rooibos Tea is rather limited.' Changing consumer attitudes and the focus on 'anti-ageing' soon afterwards placed rooibos in a position to exploit its health-promoting properties, and particularly its antioxidant activity in marketing (Wilson, 2005).

Japanese researchers were the first to study the antioxidant and 'anti-ageing' properties of rooibos. A cartoon was published in March 1992 in the Landbouweekblad, a weekly farmers' magazine, after much had been made locally of the 'anti-ageing' effect of rooibos on the skin (Fig. 6). In the cartoon Rooibos Tea Board stalwarts, J. van Putten, Deputy General Manager (Export Promotion), dressed as a nanny and pushing P. Saayman, the General Manager, in a baby stroller, calls to the flabbergasted secretary to phone Japan to inform them that the 'anti-ageing' effect had worked. The logo of the Rooibos Tea Board, symbolising a cup of rooibos tea with steam rising, forms the baby stroller. Since then many studies on the potential health-promoting properties of rooibos have been carried out in South Africa and Europe (Joubert et al., 2008a). The latest studies are summarised in Table 5.

Human studies dealing with the health promoting aspects of rooibos and the bioavailability of rooibos flavonoids, especially aspalathin, have been limited to date and are summarised in Joubert et al. (2008a) and Tables 5,6 and 7. For a compound to exert an effect in vivo, it should reach the target tissue(s). In the first instance the presence of the compound in the plasma or urine is essential. Since the study of the bioavailability of aspalathin in the pig as model (Kreuz et al., 2008), new insights into the bioavailability of aspalathin and other rooibos flavonoids have been gained by the studies of Courts and Williamson (2009), Stalmach et al. (2009) and Breiter et al. (2011). Aspalathin is extensively metabolised and phase II metabolites have been found in the urine (Table 6). Most of the metabolites were excreted in the urine of volunteers within 5 h of consuming 500 mL of rooibos ready-to-drink beverages (Stalmach et al., 2009). Kreuz et al. (2008), feeding pigs a high concentration of aspalathin-enriched green rooibos extract, found only trace quantities of the unmetabolised compound in the plasma. Its presence was recently confirmed in human plasma (Breiter et al., 2011). All of the studies showed poor

Table 6

	Summary of studies on	in vitro and	in vivo	absorption,	metabolism and	l transport of r	cooibos flavonoids
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Characteristic	Test sample	Model	Outcome	Reference
Transport across skin	Aspalathin-enriched unfermented extract; aspalathin	Human abdominal skin in Franz diffusion cells	Low level of permeation through skin and distribution in skin layers	Huang et al., 2008b
Transport across intestinal epithelial cells	Aspalathin-enriched unfermented extract; aspalathin	Caco-2 cell monolayers	Fast, concentration-dependent transport of aspalathin; higher transport from extract than pure aspalathin	Huang et al., 2008b
Absorption and metabolism	Aspalathin-enriched unfermented extract	Oral administration to pigs over an 11 day period	Aspalathin and five metabolites ( <i>O</i> -methylated and/or glucuronidated aspalathin, a glucuronidated aglycone of aspalathin and a <i>O</i> -methylated and glucuronidated eriodictyol metabolite) identified in urine; no metabolites detected in plasma	Kreuz et al., 2008
Hepatic and intestinal methylation	Aspalathin	Metabolism in rat liver and intestinal cytosolic fractions with cofactors	Two methylated metabolites identified with higher rate for intestinal cytosolic fraction	Courts and Williamson, 2009
Absorption and metabolism	Unfermented aq. drink	Single dose oral administration to humans	One methylated and one methylated-glucuronidated metabolite identified in urine	Courts and Williamson, 2009
Absorption and metabolism	Unfermented and fermented aq. extracts in ready-to-drink beverage	Single dose oral administration to humans	Eight metabolites ( <i>O</i> -linked methyl, sulfate, and glucuronide metabolites of aspalathin and an eriodictyol- <i>O</i> -sulfate) identified in urine; no metabolites detected in plasma	Stalmach et al., 2009
Hepatic metabolism	Aspalathin; nothofagin	Metabolism in rat liver microsomal and cytosolic fractions with cofactors	Two aspalathin and two nothofagin monoglucuronidated metabolites observed; one sulphated aspalathin metabolite observed; no sulphated nothofagin metabolites observed; aspalathin glucuronidated metabolites had no antioxidant activity in HPLC-ABTS <sup>++</sup> or -DPPH <sup>+</sup> scavenging assays	Van der Merwe et al., 2010
Absorption and metabolism	Unfermented aq. drink and aspalathin-enriched fraction dissolved in water	Single dose oral administration to humans	Six aspalathin metabolites (methylated, glucuronidated, methylated-glucuronidated and sulphated), three aspalathin aglycone metabolites (glucuronidated), one nothofagin metabolite (glucuronidated), as well as unmetabolised aspalathin and nothofagin, detected in urine; aspalathin, orientin, isoorientin, ( <i>S</i> )- eriodictyol-8- <i>C</i> -glucoside, vitexin and an isomer of rutin detected unmetabolised in plasma	Breiter et al., 2011

Table 7

$\sim$	N	- C	•	•. 1	•	•	. 1*		.1	1	• .	- C	.1	•	•1	0	• •		1 .1	•	• .•	1	• •	
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Bioactivity/ pharmacological property	Model	Outcome	Reference
Aspalathin			
Anti-mutagenicity	<i>Salmonella typhimurium</i> assay; tester strain/mutagen combinations TA98/2-AAF and TA100/AFB <sub>1</sub> ; with and without metabolic activation	Moderate anti-mutagenic effect; no clear dose-response effect; no mutagenic or comutagenic effect	Snijman et al., 2007
Hypoglycaemic activity	Cultured L6 myotubes	Increased glucose uptake	Kawano et al., 2009
·	RIN-5F pancreatic β-cells	Increased insulin secretion	Kawano et al., 2009
	Oral administration to db/db mice	Dietary intake supressed fasting blood glucose levels; improved glucose tolerance	Kawano et al., 2009
Hypoglycaemic activity	STZ-induced hyperglycaemic Wistar rats	Aspalathin in combination with rutin, but not alone, lowered blood glucose levels	Joubert et al., 2010a,b
Phytoestrogenicity	ECOLOGIENA® Estrogen (E1/E2/E3) ELISA kit	Moderate phytoestrogenicity	Shimamura et al., 2006
Orientin			
Anti-mutagenicity	Salmonella typhimurium assay; tester strain/mutagen combinations TA98/2-AAF and TA100/AFB <sub>1</sub> ; with and without metabolic activation	Typical dose-response anti-mutagenic effect against AFB <sub>1</sub> ; effect against 2-AAF similar to that of aspalathin; no mutagenic or comutagenic effect	Snijman et al., 2007
Anti-adipogenesis activity	3T3-L1 mouse adipocytes	Inhibited adipogenesis; decreased C/EBP $\alpha$ and PPAR $\gamma$ protein expression; vitexin also active	Kim et al., 2010
Inhibition of adrenal steroidogenic P450 enzymes	COS1 cells expressing baboon CYP21	Inhibited CYP21	Perold, 2009
Isoorientin			
Anti-mutagenicity	<i>Salmonella typhimurium</i> assay; tester strain/mutagen combinations TA98/2-AAF and TA100/AFB <sub>1</sub> ; with and without metabolic activation	Typical dose-response anti-mutagenic effect against AFB <sub>1</sub> ; more effective against 2-AAF than aspalathin; no mutagenic or comutagenic effect	Snijman et al., 2007
Hypoglycaemic and anti- hyperlipidaemic activity	Subacute oral administration to STZ-induced diabetic rats	Reduced fasting blood glucose, cholesterol and triglyceride levels	Sezik et al., 2005
Anti-inflammatory activity	Oral administration in mice; carageenan-induced pleurisy model	Inhibited leukocytes and neutrophils	Zucolotto et al., 2009
Anti-inflammatory activity	Single oral dose in mice; carrageenan-induced hind paw edema model	Inhibited inflammation without inducing gastric damage	Küpeli et al., 2004
Anti-nociceptive activity	Single oral dose in mice; <i>p</i> -benzoquinone-induced abdominal constriction model	Reduced number of writhings	Küpeli et al., 2004
Gastroprotective	Single oral dose in mice; EtOH-induced ulcerogenesis model	No activity	Küpeli et al., 2004
Hepatoprotective activity	CCl <sub>4</sub> -induced hepatotoxicity in rats	Reduced MDA and GSH levels in tissue and MDA, ALT and AST levels in plasma	Deliorman Orhan et al., 2003

2-AAF, 2-acetamido-fluorene; AFB<sub>1</sub>, aflatoxin B<sub>1</sub>; ALT, alanine aminotransferase; AST, aspartate aminotransferase; C/EBP, CCAAT-enhancer-binding protein; EtOH, ethanol; GSH, reduced glutathione; MDA, malondialdehyde; PPAR, peroxisome proliferators-activated receptor; STZ, streptozotocin.

bioavailability for aspalathin, yet despite this, it's in vivo bioactivity, e.g. glucose-lowering effect, confirm the importance and relevance of this rooibos flavonoid (Table 7).

The finding that the plasma antioxidant status of the volunteers drinking the ready-to-drink rooibos beverage peaked after 1 h (Villaño et al., 2010) may explain why Sauter (2004) showed no improvement in the plasma antioxidant status of volunteers dosed with aspalathin-enriched extract. In the latter study, the volunteers received a twice daily dose of 250 mg of aspalathin-enriched green rooibos extract for 2 weeks. In this case the blood was sampled the day after the last dose. Conjugation of aspalathin would also impair its ability to act as antioxidant in the plasma (Van der Merwe et al., 2010).

Marketing of rooibos as an 'antioxidant' beverage may need reviewing because of the over-supply of natural antioxidant products on the market. The market is constantly searching for new products that have more to offer than just high antioxidant activity and modulation of oxidative stress. Markets are moving towards condition-specific antioxidants, especially those aimed at inflammation (Crawford, 2011).

Two incorrect nutritional 'facts' that need highlighting and that some companies use in their promotional literature and on their web pages, are the erroneous claims that rooibos has a high ascorbic acid and mineral content. Hesseling et al. (1979) incorrectly interpret the apparently high ascorbic acid content of the infusion obtained using a method that is not specific for

ascorbic acid (Joubert et al., 2008a). Unfortunately, Morton (1983) repeated this high value in her review paper on rooibos, an incorrect statement which even now is still recurring in some promotional material. Furthermore, the same paper by Morton (1983) reported highly inflated values for the iron, calcium, magnesium, phosphate and potassium content of rooibos. Statements about the 'isotonic' value of rooibos have also been made. This has not been challenged in the scientific literature until recently. Joubert et al. (2008a) reported that a 5min infusion of rooibos at 'cup-of-tea' strength contains only traces of iron, with the iron content only 15% of that reported by Morton (1983). The content of other minerals was also substantially lower than previously reported. Malik et al. (2008), after determining the mineral content of a 15-min infusion reported similar values to Joubert et al. (2008a). They came to the conclusion that rooibos is not a valuable source of nutrient minerals as claimed in promotional material of some marketing companies. With the establishment of SARC, dissemination of new findings relating to the potential healthbenefits of rooibos is handled in a responsible manner. Yet, in spite of these efforts by SARC, inflated claims about this herbal tea still abound in promotional material of companies.

## 10. Future challenges and opportunities

The future challenges and opportunities of the rooibos industry are succinctly summarised by the following conclusion reached by the recent Rooibos and Honeybush Market Development Programme Framework report (Anon, 2010) after inputs of various industry stakeholders: 'An integrated global marketing drive based on the scientific proof of health claims, together with innovative, value-added product applications and a sound pricing system, could be the beginning of an extraordinary bright future for the Rooibos industry'.

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