Original article:
The Effects of *Aspalathus linearis* (Rooibos Tea) on Nitric Oxide (NO) and Cytokine Activity

*Mujeeb Hoosen*

**Abstract**

African plants have been used for medicinal purposes for many centuries. Many of these African medicinal plants are assumed to be safe but have yet to be scientifically validated. *Aspalathus linearis* (rooibos) is a commercialised South African tea recognised for its phytopharmaceutical potential. *Aspalathus linearis* (rooibos) has been gaining popularity globally for its health benefits and accepted as a nutraceutical due to the growing evidence of its efficacy. The bioactive constituents found in *Aspalathus linearis* (rooibos) have been reported to exert both anti-inflammatory and antioxidant activity however a few *in vitro* studies has suggested otherwise. *Aspalathus linearis* (rooibos) has shown to modify the actions of the immune system by influencing the regulation of messenger molecules like cytokines and nitric oxide however most of these studies have been conducted *in vitro* with a very few studies reaching *in vivo* application. Divergent *in vitro* cell models has shown to produce varying results regarding cytokine and nitric oxide NO activity of *Aspalathus linearis* (rooibos). This review highlights recent studies on the (NO) and cytokine activities of *Aspalathus linearis* (rooibos) both *in vitro* and *in vivo*. Most studies report on its anti-inflammatory and antioxidant activity however a few *in vitro* studies suggests opposite effects which should be considered for prolonged use especially when prescribed in a supplementation form. Many studies have looked at aspects of safety and toxicity of *Aspalathus linearis* (rooibos) however no complete toxicological studies have been done as yet.

**Keywords:** *Aspalathus linearis*, rooibos tea, nitric oxide, cytokine activity

**Introduction**

In recent years there has been a dramatic increase in the use of herbal medicine around the world. Herbal medicine refers to plants, parts of plants or extracts from plants that are used in health care or disease prevention. The World health organization (WHO) confirms that herbal medicine has an important role to play in the healthcare of many developing countries.

In Africa, plants have been used for medicinal purposes for many centuries. Herbal medicine plays an important role in many forms of complementary and traditional medicine systems such as African traditional medicine and Unani-Tibb, amongst others which are practiced and utilized by millions. Many African people rely on herbal medicines for their healthcare needs. Most of the western world views African herbal medicine as mysterious due to the lack of scientific evidence regarding safety and efficacy.

Herbal medicine displays distinctive characteristics that are often not well understood. The bio-activities are mostly unknown, with supportive evidence for safety and efficacy very rare. Since these herbal medicines are easily accessible over the counter and at times cheaper than conventional drugs, many patients use self-prescribed herbal medicine without revealing this to their healthcare practitioners. Regulatory studies are crucial to provide data on the toxicology, pharmacodynamics and pharmacokinetics of these herbal medicines. The effectiveness and safety of many African herbal medicines has yet to be

1. Coordinator of Unani Tibb, School of Natural Medicine, Faculty of Community and Health Sciences, University of the Western Cape, South Africa.

**Correspondence to:** Dr Mujeeb Hoosen, School of Natural Medicine, Faculty of Community and Health Sciences, University of the Western Cape, South Africa.

*Email:* mahoosen@uwc.ac.za / mujeebh786@gmail.com
established scientifically\(^4\). Recent findings have shown that up to 25\% of higher plants of the world are found south of Saharan Africa. Over 5000 African medicinal plants have already been identified, with over 16,300 medicinal uses. *Aspalathus linearis* is a popular herbal tea (rooibos) used extensively in South Africa as well as in other African regions. It is gaining popularity the world over and has major economic and therapeutic value\(^5\).

*Aspalathus linearis* (rooibos) has been reported to have anti-inflammatory properties however there are a few studies that are conflicting\(^6\). *In vitro* and *in vivo* study observations have shown that herbal medicine may exert suppression or enhancement of immune functions\(^7\). The bioactive constituents found in herbal medicine, like plant phenols, vitamins, carotenoids, phytoestrogens and terpenoids have been shown to alter various functions of inflammatory cells and these herbal constituents may affect the immune system directly or indirectly\(^8\).

Herbal medicines may modify the actions of the immune system by influencing the regulation of messenger molecules like cytokines, nitric oxide, hormones, neurotransmitters, and other peptides\(^9\). Cytokines are widely researched for many inflammatory and immune related diseases however the majority of the investigations of herbal medicine and cytokine activity have been conducted via *in vitro* assays\(^9\). The most popular cytokines assessed were IL-4, IL-6, IL-10, TNF and IFN-g. Pro-inflammatory cytokines like TNF-a, IL-1, IL-6, IL-8 and NO have been implicated in numerous immunological conditions\(^8\). The majority of *in vitro* experiments in herbal medicine make use of aqueous extractions as opposed to ethanolic extractions with varying concentrations of the wet or dried plants ranging between 0.3ug/ml to 1g/ml. Popular cell types used are splenocytes, T cells, monocytes and macrophages and the most common stimulants utilized for immune pathway activations are Con A, phytohemagglutinin (PHA) and LPS. *In vitro* cell culture incubation times varied from 6 hours to 4 weeks\(^8\).

The overproduction of NO is responsible for inflammation in several pathophysiological conditions like cancer, rheumatoid arthritis, diabetes, liver cirrhosis and septic shock. Inhibition of NO has become the main focus area in the field of anti-inflammatory research\(^10\). Herbal medicines may be valuable in the modulation of NO. iNOS is a popular investigated enzyme system utilised for *in vitro*, *ex vivo*, *in vivo*, animal, or human research on herbal medicine. Research on herbal medicines in whole, standardized or extract forms are frequently investigated with regards to nitric oxide activity\(^11\).

This paper highlights recent studies on the nitric oxide (NO) and cytokine activities of *Aspalathuslinearis*(rooibos) both *in vitro* and *in vivo*.

**An overview of *Aspalathus linearis*(rooibos) tea:**

*Aspalathus linearis* (rooibos) tea is a commercialized popular health drink from South Africa. It is most commonly used as a beverage and in cosmetic products. It is a 1.5 meter high shrub consisting of bright green needle-shaped leaves and pea-shaped yellow flowers\(^12\). *Aspalathus linearis* (rooibos) tea is considered to be one of the ‘big five’ phytomedicines in South Africa due to its popularity as an herbal health drink. A traditional beverage consumed by the Khoi people, *Aspalathus linearis* (rooibos) originates from the Cederberg area in the Western Cape\(^13\).

*Aspalathus linearis* (rooibos) tea also known as Redbush tea has several health benefits which includes antioxidant effect. Due to its low tannin content, zero caffeine and potent antioxidant properties, *Aspalathus linearis* (rooibos) has been gaining popularity globally as much more than a health beverage and accepted as a nutraceutical\(^14\). The potent antioxidant action of *Aspalathus linearis* (rooibos) tea has been attributed to its rich and unique polyphenol content\(^15\). *Aspalathus linearis* (rooibos) has shown no cytotoxic effects at all concentration tested *in vitro* using whole blood cell cultures. *Aspalathus linearis* (rooibos) has gained popularity globally as an accepted nutraceutical. The health-promoting benefits of *Aspalathus linearis* (rooibos) have been confirmed in several *in vitro* and *in vivo* studies\(^16\). There are consistent reports on the safety of *Aspalathus linearis* (rooibos)\(^6\).

**The chemical composition of *Aspalathus linearis* (rooibos) tea:**

*Aspalathus linearis* (rooibos) is known to contain various flavonoids which include flavonols, flavones and dihydrochalcones. Aspalathin, a C-C linked dihydrochalcone glucoside is considered to be the main flavonoid in *Aspalathus linearis* (rooibos)\(^17\). Other known flavonoids isolated from *Aspalathus linearis* (rooibos) are; aspalalinin, nothofagin, orientin, iso-orientin, isovitexin, dihydro-orientin, dihydro-isoorientin, isovitexin, dihydro-orientin, dihydro-isoorientin,
Aspalathus linearis (rooibos) extracts have been suggested to have anti-diabetic effects. Others claim that aspalathin can be used as a medicament for the treatment of neurological and psychiatric conditions of the central nervous system. Studies support the potential healing role of Aspalathus linearis (rooibos) in skin conditions. A comparative study on the chemo protective properties of various herbal teas in rat models revealed that Aspalathus linearis (rooibos) significantly (p<0.05) inhibits cancer promotion induction of fumonis B1 in rat liver. 

Antioxidant studies on Aspalathus linearis (rooibos) tea:

An antioxidant is a substance that can trap free radicals before oxidative damage occurs. Antioxidants such as flavanoids, polyphenol and phenolic acids are able to scavenge free radicals thereby preventing oxidative cellular damage. The link between oxidative stress and inflammation is well known especially with regards to stress related chronic diseases and accelerated aging. Two in vitro studies investigated the free radical scavenging activity of Aspalathus linearis (rooibos) and its effect on reactive oxygen species (ROS), catalase (CAT), and superoxide dismutase (SOD). The oxygen radical absorbance capacity (ORAC) assay was used to quantify the antioxidant capacity of Aspalathus linearis (rooibos) in a cancer and diabetic model using human umbilical vein endothelial cells (HUVECs) and HeLa cells. The results showed statistically significant decreases (p<0.05) in the fluorescence intensities as compared to the control. This study noted that even though antioxidant effects of Aspalathus linearis (rooibos) were observed caution should be practised for in vivo application as previous studies have reported on pro-oxidant effects of the phenolic compound in Aspalathus linearis (rooibos). 

A controlled clinical trial based on a 12 week pre-measurement and post-measurement single group intervention design was conducted to assess the effects of Aspalathus linearis (rooibos) on oxidative stress and biochemical parameters in adults at risk for cardiovascular disease. Total polyphenols, lipid peroxidation (conjugated dienes-CDS, thiobarbituric acid reactive substances-TBARS), redox status (total glutathione-GSH, ratio of reduced to oxidized glutathione-GSH:GSSG), lipid profile (total cholesterol, low density lipoprotein-LDL and high density lipoprotein-HDL cholesterol and triacylglycerol-}

hemipholphrin, quercetin, quercetin-3-robinobioside, hyperoside, isoquercetrin and rutinamoust others. Aspalathus linearis (rooibos) also contains non-flavonoid components such as lignans (vladinol E, secoisolauri resinol, secoisolauri resinol glucoside) and phenolic acids (cafeic acid, ferulic acid, p-coumaric acid, p-hydroxybenzoic acid, vanillic acid, protocatechuic acid)17,18. 

The medicinal value of Aspalathus linearis (rooibos) tea:

The absence of alkaloids and its low tannin content accounts for the reason why many consider Aspalathus linearis (rooibos) tea to be harmless. In 1968 Aspalathus linearis (rooibos) was recognized as an anti-colic agent as it relieved vomiting and chronic restlessness in babies. The anecdotal uses suggest that Aspalathus linearis (rooibos) exerts anti-allergic, appetite stimulatory and sedative effects17. Several compounds in Aspalathus linearis (rooibos) tea have been recognized to have antioxidant activity namely; aspalathin, iso-orientin, orientin, rutin, isovitexin, vitexin, isoquercitrin, hyperoside, quercetin, luteolin and chryssoeroyl. Aspalathus linearis (rooibos) revealed low anti-microbial activity when compared to other herbal tea infusions. Aspalathin, isoorientin, orientin and rutin are the compounds that are suggested to contribute to the anti-microbial activity of Aspalathus linearis (rooibos)19. In one in vitro study the anti-mutagenic effects of Aspalathus linearis (rooibos) were attributed to the flavonoid compounds aspalathin, nothofagin, luteolin and chrysoeroyl. Various studies have confirmed the anti-mutagenic, antioxidant and dermatological benefits of Aspalathus linearis (rooibos) extracts20.

Aspalathus linearis (rooibos) had no effects on the metabolic activity of LPS stimulated RAW 264.7 cells at all concentration tested indicating that this herbal extract is non-toxic even at high concentrations. The popular use of Aspalathus linearis (rooibos) over time has contributed to the assumption of its relative safety. Many studies have looked at aspects of safety and toxicity of Aspalathus linearis (rooibos) however no complete toxicological studies have been done as yet.

One study suggested that the minor component of Aspalathus linearis (rooibos) quercetin, is implicated in its possible mutagenic effects. However these effects were seen in concentration of 220-230 times more than that of the normal tea drinking quantities15. Aspalathin found in certain
levels) as well as liver and kidney function were included. This study reported a decreased lipid profile (decreased HDL and saturated fats) and redox status in the participants however there were no significant changes to the non-specific marker of generalized inflammation, C-reactive protein, after the 6 week consumption of *Aspalathus linearis* (rooibos) extract.

An *in vivo* study used *Caenorhabditis elegans* as a model organism to assess the effect of *Aspalathus linearis* (rooibos) extracts against oxidative stress. This study employed juglone, a generator of ROS, known to cause damage to cells and organisms. Age-synchronized *Caenorhabditis elegans* were treated with green *Aspalathus linearis* (rooibos) extract (100µg/ml), red *Aspalathus linearis* (rooibos) extract (100µg/ml) or aspalathin (0, 10, 20 and 50µM). After exposure to acute oxidative damage, the surviving organisms were examined and scored. *C. elegans* treated with *Aspalathus linearis* (rooibos) extract displayed an extended lifespan. Green *Aspalathus linearis* (rooibos) extract exhibited a more potent antioxidant effect than red *Aspalathus linearis* (rooibos), most likely due to its higher aspalathin content. Quantitative real-time PCR results demonstrated that aspalathin reduced endogenous intracellular level of ROS by targeting stress and ageing related genes. This study suggests that the antioxidant effect of *Aspalathus linearis* (rooibos) could be mediated by regulation of the DAF-16/FOXO insulin-like signalling pathway.

The potent antioxidant action of *Aspalathus linearis* (rooibos) tea has been attributed to its rich and unique polyphenol content. Flavonoids (luteolin and quercitin) in *Aspalathus linearis* (rooibos tea) showed inhibition of pro-inflammatory cytokines, IL-6 and TNF-α using a LPS-stimulated macrophage model. One study on the water extract of *Aspalathus linearis* (rooibos) in murine splenocytes using anti-ovalbumin (OVA) and sheep red blood cells showed an increase in antibody production with no effect seen in lipopolysaccharide-stimulated (LPS) spleen cells. *Aspalathus linearis* (rooibos) stimulated interleukin (IL)-2 in splenocytes primed with OVA and CD3, whilst down regulating IL-4 in OVA primed cells. Similar results were seen *in vivo*, in Wistar rats given oral doses of *Aspalathus linearis* (rooibos) water extract. These cyclosporine treated rats displayed restoration in OVA-induced antibody production however no difference was seen between OVA-stimulated and control rats. In a previous study, Joubert et al., reported on both antioxidant and/or pro-oxidant activities of the aqueous extracts and crude polyphenolic fractions of unfermented and fermented *Aspalathus linearis* (rooibos). These results are contrary to several *in vitro* and *in vivo* reporting on the antioxidants and/or anti-inflammatory effects of *Aspalanthuslinearis* (rooibos) as previously mentioned. Some *in vitro* studies used the ethanolic extract of *Aspalanthuslinearis* (rooibos) which may also account for differences in findings. The potential adverse biological effects of the *Aspalathuslinearis* (rooibos) extracts should be considered when used as a dietary supplement.

**Anti-inflammatory studies on *Aspalathus linearis* (rooibos) tea**

Active oxygen and free radicals can induce inflammation. Vitamin C and E, flavonoids and enzymes such as catalase, glutathione peroxidase (GPx), and serum superoxide dismutase (SOD) are known antioxidants. An *in vivo* study investigated the anti-inflammatory effects of *Aspalathus linearis* (rooibos) tea using a rat colitis model. SOD levels were determined as an indicator of antioxidant activity whilst urine 8-hydroxy-2′-deoxyguanosine (8-OHdG) concentrations were indicative of DNA damage. Dextran sodium sulfate (DSS) was used to induce colitis in rodents. Clinical symptoms, hemoglobin, serum iron, (8-OHdG) concentration and SOD levels were compared between the *Aspalathus linearis* (rooibos) and control groups. Levels of SOD of the *Aspalathus linearis* (rooibos) group were significantly increased (P<0.05) compared with the controls whilst urine 8-OHdG levels were significantly decreased (P<0.05) in the *Aspalathus linearis* (rooibos) group compared with the controls. The antioxidant activity of *Aspalathus linearis* (rooibos) tea were proposed as the mechanism preventing DNA damage and inflammation. *Aspalathus linearis* (rooibos) was included in an anti-inflammatory study using LPS-stimulated macrophage model. Several herbs and spices and their components were included in this study. This study reported a 25% reduction in IL-6 secretion in samples incubated with *Aspalathus linearis* (rooibos) (0.5mg/ml) and
a significant reduction in IL-10 as well. Western blot analysis showed an increased expression of COX-2 (more than 25%) by the extracts of Aspalathus linearis (rooibos) tea (0.5mg/ml)\textsuperscript{26}. A study on the effect of Aspalathus linearis (rooibos) on rat adrenal cytokine expression showed that Aspalathus linearis (rooibos) decreased IL-6 synthesis and increased IL-10 production in cortical tissue\textsuperscript{27}. An in vitro and in vivo study assessed the effects of the two main actives dihy-drochalcones (aspalathin and nothofagin) of Aspalathus linearis (rooibos) on high glucose-mediated vascular hyperpermeability, adhesion of monocytes toward HUVECs and glucose-mediated vascular hyperpermeability, adhesion of monocytes toward HUVECs and glucose-mediated vascular hyperpermeability, adhesion of monocytes toward HUVECs and glucose-mediated vascular hyperpermeability. Aspalathus linearis and nothofagin inhibited high glucose-induced inflammation using human umbilical vein endothelial cells (HUVECs) and mice. Aspalathin and nothofagin inhibited high glucose-mediated vascular hyperpermeability, adhesion of monocytes toward HUVECs and expression of CAMs in a dose dependant manner whilst significantly suppressing ROS formation (P<0.05) and NF-κβ activation (P<0.05)\textsuperscript{28}. An in vitro study showed that the inflammatory potential at these concentrations in unstimulated RAW 264.7 cells. This suggests that Aspalathus linearis (rooibos) possess pro-inflammatory potential at these concentrations in absence of a stimulus in vitro. This study noted that Aspalathus linearis (rooibos) is mainly consumed as a health promoting beverage over prolonged periods and therefore its pro-inflammatory potential should be considered in vivo especially in chronic inflammatory conditions as NO stimulation is responsible for cellular and tissue damage which contributes to numerous inflammatory conditions affecting different organs. Mueller et al.,conducted a similar study on Aspalathus linearis (rooibos) on RAW 264.7 macrophages as mentioned earlier. Results showed decreased IL-6 at concentrations of 500µg/ml and 1000µg/ml in unstimulated RAW 264.7 cells. This suggests that Aspalathus linearis (rooibos) possess pro-inflammatory potential at these concentrations in absence of a stimulus in vitro. This study noted that Aspalathus linearis (rooibos) is mainly consumed as a health promoting beverage over prolonged periods and therefore its pro-inflammatory potential should be considered in vivo especially in chronic inflammatory conditions as NO stimulation is responsible for cellular and tissue damage which contributes to numerous inflammatory conditions affecting different organs. Mueller et al.,conducted a similar study on Aspalathus linearis (rooibos) on RAW 264.7 macrophages as mentioned earlier. Results showed decreased IL-6 at concentrations of 0.5mg/ml\textsuperscript{31}. Differences in concentration between (0-1000 ug/ml) however within different models which may account for variations in findings. Variations in the use of ethanolic and aqueous extracts in previously mentioned studies may also account for differences in findings due to the presence of different bio-actives in aqueous extracts compared to ethanol extracts.Waisundara and Hoon’s study also cautioned on the in vivo application of these in vitro pro-oxidant reports of Aspalathus linearis (rooibos) as mentioned previously\textsuperscript{16}. Most studies reports on the anti-inflammatory effects of Aspalathus linearis (rooibos) however, there are a few studies which reports on its pro-inflammatory effects\textsuperscript{9}. 

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### Table 1. Studies on the effects of Aspalathus linearis (rooibos) on cytokine and NO activity

<table>
<thead>
<tr>
<th>Preparation</th>
<th>Dose</th>
<th>Model</th>
<th>Cytokine and/or NO effect</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.63g leaves/100ml water, boiled for 15min and freeze-dried</td>
<td>1-1000µg/ml</td>
<td><em>in vitro</em>, murine splenocytes</td>
<td>Increased IL-2, Inhibition of IL4 (OVA-primed murine splenocytes)</td>
<td>Kunishiro et al., 2001</td>
</tr>
<tr>
<td>1.63g leaves/100ml water, boiled for 15min</td>
<td>4ml/day extract for 3 weeks</td>
<td><em>ex vivo</em>, splenocytes of female mice</td>
<td>Increased IL-2 (OVA-induced)</td>
<td>Kunishiro et al., 2001</td>
</tr>
<tr>
<td>5g/100 ml freshly boiled phosphate-buffered saline, steeped for 10 min</td>
<td>0-730µg/ml</td>
<td><em>in vitro</em>, cultured human umbilical veins endothelial cells</td>
<td>Increased NO production</td>
<td>Persson et al., 2006</td>
</tr>
<tr>
<td>17.5g/1000ml boiled for 15mins</td>
<td>100µg/ml in vitro</td>
<td><em>in vitro</em>, murine splenocytes</td>
<td>Increased IL-10 (OVA-induced) Decreased IL-2, IL-4 IFNγ</td>
<td>Ichiyama et al., 2007</td>
</tr>
<tr>
<td>25g tea bags/1000ml Seeped in boiled water, freezeed</td>
<td>0-250µg/ml</td>
<td><em>in vitro</em>, whole blood culture</td>
<td>Increased IL-6, IL-10, and IFNγ (unstimulated) Increased IL-6, decreased IL-10 and No effect on IFNγ (LPS/PHA stimulated)</td>
<td>Hendricks and Pool, 2010</td>
</tr>
<tr>
<td>10g tea in 400ml fresh-boiled water for 5 min</td>
<td>400ml per week for 4 weeks</td>
<td><em>in vivo</em>, randomized three-phase crossover design (human)</td>
<td>No effect on NO</td>
<td>Persson et al., 2010</td>
</tr>
<tr>
<td>100mg/ml DMSO powder leaf extract</td>
<td>0.5mg/ml</td>
<td><em>in vitro</em>, RAW 264.7 macrophages</td>
<td>Decreased IL-6 and IL-10 (LPS-stimulated) No effect on TNFα</td>
<td>Mueller et al., 2010</td>
</tr>
<tr>
<td>15g leaves/250ml (human) 160mg dried extract/ml (rat)</td>
<td>90g <em>Aspalathus linearis</em> (rooibos) leaves/subject daily 0.25g <em>Aspalathus linearis</em> (rooibos) leaves/ rat daily</td>
<td><em>in vivo</em>, humans and rats</td>
<td>Increased IL-10 inhibition of IL-6</td>
<td>Swart et al., 2013</td>
</tr>
<tr>
<td>2g/100 ml, seeped for 30mins</td>
<td>2%, w/v</td>
<td><em>in vivo</em>, rats</td>
<td>Decreased TNFα and IL-6 No effect on IL-1β and IL-10</td>
<td>Ajewon et al., 2014</td>
</tr>
<tr>
<td>Aspalathin and Nothofagin, <em>Aspalathus linearis</em> (rooibos) compounds</td>
<td>10μM-30μM 8.7µg- 27.1µg (per mouse)</td>
<td><em>in vitro</em>, human umbilical vein endothelial cells</td>
<td>Decreased TNFα and IL-6</td>
<td>Lee and Bae, 2015</td>
</tr>
<tr>
<td>30g plant (rooibos) material with 300ml chloroform for 8h using a glass soxhlet in 300ml methanol.</td>
<td>twice daily by gavage (250µl), with a gavage dosage extracted from 0.25g rooibos leaves</td>
<td><em>in vivo</em>, rats</td>
<td>Decrease IL-6 Increased IL-10</td>
<td>Smith and Swart, 2016</td>
</tr>
<tr>
<td>25g tea bags/500ml Seeped in boiled water, freezeed</td>
<td>0-1000µg/ml</td>
<td><em>in vitro</em>, RAW 264.7 macrophages</td>
<td>Increased IL-6 and NO (unstimulated) No effect on IL-6 and NO (LPS stimulated)</td>
<td>Hoosen and Pool, 2018</td>
</tr>
</tbody>
</table>

*Aspalathus linearis* (rooibos) is has gained acceptance due to its common use and the growing evidence of its relative safety however there are a few studies that may suggest otherwise. Inconsistent results are a common occurrence seen when *in vitro* and *in vivo* studies are compared for herbal products.

34 This could be due to the differences in the concentrations tested. Other contributing factors to differences in findings of these studies could be attributed to variations that
exist in different batches of the tea sample. The chemical composition of herbs differ depending on various factors which includes the botanical species, the anatomical part of the plant used, storage methods, sun, humidity, type of soil, time of harvest, geographic location amongst others. Batch to batch variations can be found within the same manufacturing company which can result in significant variations in pharmacological activities influenced by pharmacodynamic and/or pharmacokinetic factors.

Aspalathuslinearis (rooibos) tea could potentially be used for prophylactic purposes. However, important consideration should be given to its possible pro-inflammatory action in midst of inflammation which could lead to or worsen tissue damage.

Conclusions

The general public makes use of herbal medicines for various reasons based on in vivo claims which includes enhancement of general wellbeing or/and for prophylactic purpose. These herbal medicines may enhance or suppress immune function which could benefit or harm patients. In depth study of the effect of herbal medicine on the immune system requires the use of both in vitro and in vivo experimentation.

In vitro studies on Aspalathuslinearis (rooibos) should be standardised and regulated in terms of dosages used. Concentrations used in vitro should be calculated based on anecdotal dosages with the objective of extrapolating in vitro data for in vivo application. This could contribute to an efficient system of researching this popular tea which could be cost effective and less time consuming. Future studies should consider that different batches of the product may produce varying results due to the complex chemistry involved. Also, there are vast pharmacological differences between aqueous and ethanol extracts of Aspalathuslinearis (rooibos) therefore studies should include both forms. Divergent in vitro cell models has shown to produce varying results regarding cytokine and NO activity of Aspalathuslinearis (rooibos).

Future research should include collaborations between complementary and traditional medicine practitioners, microbiologists and phytochemists to ensure better research outcomes. This is essential to ensure the safety and efficacy of Aspalathuslinearis (rooibos).

Conflict of interest: None declared

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