Humarine substances are effective in the suppression of delayed type hypersensitivity, rat paw oedema, a graft-versus-host reaction and contact hypersensitivity in rats. They reduce the C-reactive protein levels of patients suffering from osteoarthritis of the knee and the wheel and flare reaction of patients suffering from hay fever. They have also been described as cardioprotective and pro-angiogenic. Toxicity studies have indicated that potassium humate is safe in humans up to a daily dosage of 1 g/kg, whereas fulvic acid is safe in humans up to a daily dosage of 1.8 g per adult. The antiinflammatory action of potassium humate can be contributed to the inhibition of the release of inflammatory-related cytokines, an adhesion molecule, oxidants and components of the complement system. Copyright © 2015 John Wiley & Sons, Ltd.

**Keywords**: inflammation; humic substances; humic acid; potassium humate; fulvic acid.

**INTRODUCTION**

Long before chemical formulae to cure diseases were developed, humans discovered the healing powers of plants. Humic substances are formed from the decomposition of plants and occur naturally in water, peat, soil and brown coal. These substances have a complex structure and can be fractionated into humin and humic and fulvic acids (MacCarthy et al., 1979). Although they have primarily been used to stimulate plant growth, they have also been applied in the treatment of various diseases in humans. Humic acids are soluble in water only at pH values higher than 2 whereas fulvic acid is soluble in water even at low pH values. Humin, in contrast, is a mixture of materials that are insoluble under all conditions (Pena-Méndez et al., 2005). The richest source of humic acid is found in brown coal, also known as lignite, which is the 'youngest' part of coal. Two different humic substances (i.e. forest and grass humic substances) were characterised with the use of two different spectroscopic techniques, that is, diffuse reflectance infrared Fourier transform and nuclear magnetic resonance (NMR 1H) (Muscola et al., 2006). Owing to the significant differences found in the chemical compositions of the two samples, an analytical and preparative thin layer chromatography analysis was carried out and indicated the complexity of the molecular structure of potassium humate (Van Rensburg et al., 2010b).

**APPLICATION OF PEAT AND MUD PREPARATIONS FOR THE TREATMENT OF INFLAMMATION**

The pharmacological properties of products rich in humic acids, derived from peat extracts such as sapropel, Tolpa peat and mumie, have been extensively reviewed (Schepetkin et al., 2002, 2003). It has been used as a folk medicine for more than 3000 years. Diseases that have been treated include inflammatory-related and ophthalmological diseases, gastric ulcers, acute gastroenteritis, anaemia, hypercholesterolemia, dermatitis, psoriasis, hepatic and viral diseases and diseases of the gall bladder.

Peat preparations have been used as a topical treatment as well as in spas for dermatitis and psoriasis (Wolina, 2009) (Table 1). The topical application of mud and peat reduces the symptoms of patients suffering from rheumatoid arthritis, eczema and psoriasis (Codish et al., 2005; Chadzopulu et al., 2011) (Table 1). These products have been shown to increase T-cell immunity in patients infected with pulmonary tuberculosis and are also effective in the treatment of hepatic diseases and diseases of the gall bladder (Schepetkin et al., 2002). In a study by Krzeminski et al. (2005), it was shown that a Tolpa peat preparation (TPP) possesses pro-angiogenic and cardioprotective effects when administered subcutaneously in rats after the induction of myocardial infarction (Table 1).

Mud bath therapy and humate balneotherapy have improved the quality of life of patients suffering from osteoarthritis (Table 1), which is an inflammation-related disease associated with the progressive destruction of cartilage (Bellometti et al., 1997; Iubitskaia and Ivanov, 1999; Codish et al., 2005; Fraioli et al., 2011; Güngen et al., 2012; Chadzopulu et al., 2011), whereas Vysokogorskii et al. (2009) described the wound healing properties of a solution of sapropel applied topically to full-thickness planar wounds induced in Wistar rats (Table 1).

**MECHANISTIC STUDIES**

Mechanistic studies done in vitro indicated that potassium humate derived from brown coal inhibits degranulation of phagocytes (Joone and van Rensburg, 2004).
the activation and/or release of blood products associated with inflammation, such as cytokines and superoxide dismutase, and the migration and adhesion of inflammation-related cells to sites where allergic reactions and tissue damage take place (Van Rensburg and Naude, 2009). Fulvic acid also decreases the release of tumour necrosis factor alpha (TNF-alpha), but at much higher concentrations (>200 μg/ml) (Junec et al., 2009).

On the other hand, Chen et al. (2002) indicated that humic acid treatment increased the adhesion as well as the production of oxidants by stimulated neutrophils in vitro. Humic substances have also been reported to stimulate the release of pro-inflammatory cytokines such as TNF-alpha in vitro, but only in the presence of exogenous lipopolysaccharides (Junec et al., 2009), indicating that these substances should not cause inflammation under normal conditions.

Humic substances also possess antioxidant activities (Aeschbacher et al., 2012; Kučerík et al., 2008; Vašková et al., 2011) and inhibit the expression of complement receptor one and three in lipopolysaccharide-induced human umbilical vein endothelial cells through the inhibition of nuclear factor kappa B activation (Gau et al., 2000). These surface molecules play an important role during inflammation by assisting the cells to adhere to the walls of blood vessels in the vicinity of inflammatory reactions as in the case of patients suffering from autoimmune diseases (Crockard et al., 1992). The previously mentioned results were confirmed with a potassium humate product derived from brown coal (Joone and van Rensburg, 2004). In this way humic substances protect areas of existing inflammation by stopping inflammatory cells from reaching affected sites, ‘sticking’ to the nearby blood vessels and releasing toxic substances in these areas. The mechanism of action of humic substances can also contribute to the inhibition of both the classical and alternative pathways of complement activation, as well as the degranulation of phagocytes and the production of inflammation-related cytokines such as IL-1β, IL-6, IL-10 and TNF-α (Van Rensburg and Naude, 2009; Joone and van Rensburg, 2004).

### PRECLICAL TOXICITY AND EFFICACY STUDIES

The pro-angiogenic, angio-immunomodulatory as well as the cardioprotective properties of TPP, administered subcutaneously in rats was described by Tadeusz et al. (2005) (Table 1). They came to the conclusion that Tolpa peat can prevent the development of ischemic cardiomyopathy in rats.

The antiinflammatory activity of topically applied oxifulvic acid, a fulvic-acid product derived from bituminous coal, was compared with a 1% preparation of both diclofenac sodium and betamethasone in a murine model of contact hypersensitivity (Van Rensburg et al., 2001) (Table 1). In this experiment, mice were sensitised to dinitrofluorobenzene and then challenged with dinitrofluorobenzene on the dorsal surface of one ear. The mice’s inflamed ears were treated with a topical application of dinitrofluorobenzene on the dorsal surface of one ear. The mice’s inflamed ears were treated with a topical

### Table 1. A summary of the various successful trials (preclinical and clinical) done on humic substances

| Humic substances | Used by/tested in | Route of application | Application |
|------------------|------------------|----------------------|-------------|
| Peat             | Humans           | Topical and in spas  | Treatment of |
|                  |                  |                      | Dermatitis (Wolina, 2009) |
|                  |                  |                      | Psoriasis (Codish et al., 2005) |
|                  |                  |                      | Rheumatoid arthritis (Gingen et al., 2012) |
|                  |                  |                      | Wounds (Vysokogorski et al., 2009) |
| Tolpa peat preparation | Rats          | Subcutaneous        | Cardioprotective and pro-angiogenic (Krzemiński et al., 2005) |
| Mud/humate balneotherapy | Humans | Mud bath therapy | Improve quality of life (Chadzopulu et al., 2011) |
| Sapropel         | Rats             | Topical              | Wound healing (Vysokogorski et al., 2009) |
| Sapropel         | Humans           | Mud bath therapy    | Treatment of osteoarthritis (Schepeletkin et al., 2002). |
| Oxifulvic acid   | Rats             | Topical              | Wound healing (Van Rensburg et al., 2001). |
| Oxifulvic acid   | Mice             | Topical              | Inflammation (Van Rensburg et al., 2001). |
| Oxifulvic acid   | Humans           | Topical              | Inflammation (Snyman et al., 2002). |
| Carbohydrate-derived fulvic acid | Rats | Topical    | Wound healing (Sabi et al., 2011). |
| Carbohydrate-derived fulvic acid | Humans | Topical | Treatment of eczema (Gandy et al., 2011). |
| Potassium humate | Humans           | Oral                 | Decrease contact hypersensitivity (Van Rensburg et al., 2007). |
| Potassium humate | Rats             | Oral                 | Decreases delayed type hypersensitivity (Van Rensburg and Naude, 2009). |
|                  |                  |                      | Decrease graft-versus-host reaction (Van Rensburg and Naude, 2009). |
| Potassium humate |                  |                      | Decrease paw oedema and a graft-versus-host reaction (Van Rensburg et al., 2010b). |

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application of either a placebo cream or a formulation containing oxifulvic acid, diclofenac sodium or betamethasone. The thickness of their ears was measured on a daily basis. Oxifulvic acid, as well as the betamethasone and diclofenac sodium formulations, reduced the cutaneous inflammatory response.

In a preclinical toxicity study in rats treated daily with an oral dosage of potassium humate at 1 g/kg for 1 month, it was found that the dosage had no effect on the safety parameters nor did a dosage of 500 mg/kg body weight have any effect on the pups when it was administered to pregnant female rats (Van Rensburg et al., 2007). In efficacy studies, potassium humate, at an oral dosage of 60 mg/kg, inhibited a delayed type hypersensitivity reaction in rats immunised with sheep red blood cells, a carrageenan-induced oedema and a graft-versus-host reaction in rat models (Van Rensburg et al., 2010b), as well as a contact hypersensitivity reaction in rats sensitised with dinitrofluorobenzene (Van Rensburg et al., 2007) (Table 1). In these studies, potassium humate compared favourably with indomethacin and prednisolone. Interestingly, immune-incompetent rats (induced with cyclophosphamide treatment in the graft-versus-host experiment) treated with potassium humate did not suffer from the normal weight loss as was the case with rats treated with cyclophosphamide alone (Van Rensburg et al., 2010b). Furthermore, a chemical complex was prepared by Anwer et al. (2010) with humatic acid and aspirin by lyophilization. This complex enhanced the antiinflammatory activity of aspirin alone in the rat paw oedema model (Anwer et al., 2010).

The antioxidant and wound healing properties of saproplms were described by Vysokogorski et al. (2009) suggesting that it might be recommended for inclusion in medicinal formulations, whereas Ozkan et al. (2014) described the neuroprotective effects of humatic acid, administered intraperitoneally, in a focal cerebral ischemia rat model, which might be due to its antioxidant properties. They speculated that humatic acid may be applied as a preventive agent in patients with a high risk of developing ischemia-induced brain injury.

In two separate studies on two different fulvic-acid products, one derived from bituminous coal and the other from a carbohydrate source (CHD-FA), it was found that fulvic acid is safe and effective in the reduction of a contact hypersensitivity reaction in rats when applied topically (Van Rensburg et al., 2001; Sabi et al., 2011) (Table 1).

A study was done to determine the effects of the subcutaneous administration of TPP on spontaneous angiogenesis in rats after the induction of myocardial infarction. The results indicated that this product possesses cardioprotective properties by preventing the development of ischemic cardiomyopathy (Krzeminski et al., 2005) (Table 1).

Trckova et al. (2005) reviewed the application of peat as a food supplement for farm animals and came to the conclusion that there are many beneficial properties of peat such as the detoxifying and absorbent effects, stimulation of the immune system and an increase in the growth of the animals. He suggested that, because of the differences in the chemical composition of peat from different areas, it will be necessary to test the effects of each source.

**THE ANTIINFLAMMATORY PROPERTIES OF HUMIC SUBSTANCES**

### CLINICAL STUDIES

Oxifulvic acid, applied topically to allergic individuals, significantly reduced a wheel and flare reaction after intradermal allergen challenge (Snyman et al., 2002), which was similar to that of hydrocortisone (Table 1).

A pilot study done on atopic people indicated that a 4.5% oxifulvic acid cream applied topically inhibited an elicited inflammatory reaction (Gandy et al., 2011). This study was followed up with a clinical trial in which atopic people were treated for 3 days with daily oral dosages of up to 40 mL of a 3.8% solution of a carbohydrate-derived fulvic acid. A significant decrease in the skin prick test was observed (Table 1). It was concluded that this product was safe at these dosages (Gandy et al., 2012).

In a double blind placebo controlled Phase I study with oxihumate (a bituminous-coal-derived humate product) done on HIV-positive individuals at oral dosages of 2, 4, 6 and 8 g per person per day over a two-week period (Botes et al., 2002), it was demonstrated that, although the product had no positive effect on the viral load and CD4 counts of the patients, it was well tolerated with no side effects. This trial was executed before April 2004, when the national antiretroviral treatment programme was launched in South Africa.

The most conclusive findings were the results obtained from two clinical trials. In the first trial, potassium humate was administered orally in patients suffering from allergic rhinitis (Gandy et al., 2010), and in the second trial, it was administered in patients suffering from osteoarthritis of the knee (van Rensburg et al., 2010a). In the first trial, potassium humate decreased the wheel and flare reaction of the patients, and in the second trial, it improved the physical functioning of the patients and decreased the levels of C-reactive protein (CRP) in the blood of patients on the product (Fig. 1) (Table 1). This was confirmed by Güngen et al. (2012) who found that mud pack therapy slows down

![Figure 1. Effect of potassium humate (HA) versus placebo (before and after a 2 week washout period) on high sensitivity C-reactive protein (hs-CRP) levels of patients suffering from osteoarthritis of the knee**. * Significant (p < 0.5) reduction in hs-CRP compared with placebo values. **Van Rensburg C. E. J., Badenhorst B. E., Gandy J. J. and Snyman J. R., 2010b. The Open Conference Proceedings Journal 1:69–74. This is an open access article licenced under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/3.0/), which permits unrestricted, non-commercial use, distribution and reproduction in any medium, provided the work is properly cited.](http://creativecommons.org/licenses/by-nc/3.0/)
the progression of knee osteoarthritis. Although CRP is not directly involved in the inflammatory process, it is widely used as a marker of inflammation (Boylan et al., 2001; Koenig et al., 1999; Nakayama et al., 1993). For example, Nakayama et al. (1993) reported that a strong association exists between an increase in CRP levels and the progression of atherosclerosis, whereas McIntyre et al. (1997) found the measurement of CRP useful for monitoring patients suffering from inflammatory bowel disease.

**CONCLUSIONS AND FUTURE DIRECTIONS**

Inflammation plays a role during viral and bacterial infections (Nakayama et al., 1993), autoimmune diseases (Nathan, 2002), cancer (Balkwill and Mantovani, 2001; Coussens and Werb, 2002; Baumgarten and Frasor, 2012), allergies (Venge, 1994), Alzheimer disease (Holmes et al., 2009) and cardiovascular conditions (Ridker et al., 2000). An association between inflammation and malignancies has also been described (Lu et al., 2006). The effective control of inflammation could be used to protect candidates predisposed to these conditions (Abou-Raya and Abou-Raya, 2006). This could also be the case with other inflammatory-related diseases (Coussens and Werb, 2002; Halliday et al., 2000; Nakayama et al., 1993).

Unfortunately, the use of non-steroidal antiinflammatory drugs can lead to an increased risk of gastrointestinal complications such as ulcerative colitis, intestinal ulcers, intestinal perforations, damage to the small bowel (Somasundarum et al., 1995; Davis, 1995; Thieffin and Beauregerie, 2005; Sostres et al., 2010) and large intestines (Davis, 1995), as well as an increased risk of cardiovascular complications (Bjarnason et al., 1993; McIntyre et al., 1997; Fosslien, 2005). These drugs have therefore become unsafe for use by patients already predisposed to these conditions. Interestingly, a humic acid preparation (TPP) significantly accelerated the healing of gastric ulcers induced in rats (Brzozowski et al., 1994) (Abshenas et al., 2014). A chemical complex was prepared with humic acid and aspirin by lyophilisation. This complex enhanced the antiinflammatory activity of aspirin alone in the rat paw oedema model (Anwer et al., 2010).

In conclusion, products derived from humic substances have been used and tested over centuries for inflammatory-related diseases (Table 1) suggesting that it can be a possible safe alternative for the treatment and/or prevention of diseases associated with inflammation.

**Conflict of Interest**

The author is involved in the distribution of a potassium humate preparation on the market for the treatment of various inflammatory-related diseases.

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