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EFFECTS OF WASTE CRUMB RUBBER ON MEDIUM CHARACTERS AND GROWTH OF *LOLIUM PERENNE* L.

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Abstract

Crumb rubber, a product of waste rubber, may be used in sports turf medium for alleviating soil compaction. Crumb rubbers with diameter of 0.5-1 mm were applied through turfgrass rooting medium at seven scales (0%=control, 6%, 9%, 12%, 15%, 18%, 21%) and the effects of crumb rubber on turf medium characters and growth of *Lolium perenne* L., were investigated. Crumb rubber improved the properties of sand-based medium such as the decreasing bulk density and increasing water retention. Crumb rubber application didn't affect seed germination of *L. perenne* significantly as compared to control. In contrast, a significant reduction in shoot length was observed in two stubbles of *L. perenne*. However, the first stubble in 15% crumb rubber treatment and the second stubble in 12% crumb rubber treatment exhibited better growth and higher shoot length was obtained than other treatments. Addition of varying proportions of crumb rubber also markedly decreased shoot fresh and oven dry biomasses, and the effect was more pronounced at 21% crumb rubber treatment for the second stubble, but 15% crumb rubber gave the highest aboveground biomass for two stubbles among all crumb rubber treatments. There were no significant differences in the root length and fibrous root number between crumb rubber treatments and control except for 21% and 6% treatments. However, crumb rubber had positive effects on root biomass and chlorophyll content of the second stubble. Crumb rubber 15% and 18% increased root biomass by 94% and 108% respectively, relative to control. Maximum increase in chlorophyll content of the second stubble was observed at 18% crumb rubber treatment. The net photosynthetic rate (*Pn*), stomatal conductance (*Gs*) was improved at 18% proportion of crumb rubber. The findings of the trial suggested that the optimum proportion of crumb rubber added in *L. perenne* medium was between 15% and 18%, which would not affect plant growth obviously and may improve some growth parameters.

Introduction

Of the annual total global production of waste rubber material, which amounts to more than 10 million tons, approximately 60~70% is used in tyres (Lu, 2005). After they have been worn-out during their limited service life, millions of used tyres are discarded every year and hauled to a dump. In the last year, over three million tons of waste tyres were generated in the EU states (Aiello *et al*., 2009). China is a large country for rubber consumption. The annual production of waste rubber amounts to 2 million tons, 80 million scrap tires were produced in 2002, and with 12% of growth rate every year (Li, 2008; Cao, 2007). As polymeric materials, rubbers do not decompose easily. Large amounts of waste rubber materials discarded pose two major problems: wastage of valuable rubber and disposal of waste rubber leading to environmental pollution. Two major ways to solve the problem are recycling and reuse of waste rubber and reclaim of rubber raw materials (Adhikari *et al*., 2000). An attempt to reuse the abandoned tires by grinding them into small particles (crumb rubber) and use in sports field, pavement and construction materials may be an important approach to reduce waste rubbers in large quantities (Farrell, 1998; Batayneh *et al*., 2008; Ganjian *et al*., 2009; Xiao *et al*., 2009; Yilmaz & Degirmenci, 2009;). Yang *et al*., (2004) investigated the possibility of using waste tire composites reinforced with rice straw as construction materials, showing that the composite boards are able to be used as a substitute for insulation boards and other flexural materials in construction. Malmgren *et al*., (1994) stated that crumb rubber applied in race course could improve the elasticity of the track, reducing the risk of horse injuries owing to serious hardness of the field.

With the popularization and development of sports activities, the demand for sports turf such as soccer, golf and grassland tennis etc., is increasing day by day. Under long-term high-traffic condition, sports turf medium would become compaction and lose resiliency. Crumb rubber applied in sports turf as soil amendment could decrease soil compaction from heavy foot traffic (Malmgren *et al*., 1991). Studies on this aspect have mainly dealt with the effects of crumb rubber on elasticity, wear tolerance, ball roll and ball bounce of sports turf (Bowman *et al.*, 1994; Rogers *et al*., 1998). Groenevelt & Grunthal (1998) pointed out 10~20% crumb rubber significantly reduced surface hardness of sports turf. Riggle (1994) also stated that the inclusion of rubber particles in soil could improve soil physical properties such as aeration and infiltration rates of water etc., the use efficiency of water and fertilizers was improved by up to 30%. Rogers *et al*. (1998) reported that crumb rubber could alter surface characteristics and increase wear tolerance of turfgrass exposed to traffic, and the small crumb rubber was more effective. Zhao *et al*. (2009) found that the size of crumb rubber filled in turf medium was correlated with trample resistance of turf, and 1-2 mm crumb rubber was better than 2-4 mm and 4-6 mm. However, scarce data are found on responses of turfgrass growth and medium characters to crumb rubber in the literature. The aim of the present research was to investigate the effects of crumb rubber on medium characters and growth of *L. perenne*.

Materials and methods

Plant culture and treatments: Fine sand was sieved and rinsed with distilled water, then oven-dried at 80°C for about 12 h. Turfgrass medium was compounded by sand, municipal solid waste compost and crumb rubber. Crumb rubber (diameter of 0.5-1 mm) was added at seven rates of 0% (control), 6%, 9%, 12%, 15%, 18% and 21%, with 10% compost as nutrient.

Compounded medium 200 g was loaded into plastic pots (Φ = 7 cm, height = 8 cm) and 300 seeds of *L. perenne* were sown in each pot. The pots were arranged according to completely randomized design with four replications. Turfgrass culture was exposed to natural light with average temperature of 20°C and average relative humidity of 50%. Watering was performed daily throughout the study period to maintain adequate medium moisture.

Medium characters: Turf medium with different proportions of crumb rubber was determined in bulk density and total porosity using the core method described by Bao (2000) and saturated water holding capacity (Lin, 2004). The pH value was measured using the glass-electrode pH meter (pHS-3C).

Growth and biomass: Seed germination was recorded until the seedling stage. Turfgrass shoots were harvested at 38 days and 95 days after sowing for the first stubble and the second stubble respectively. Fresh shoot biomass was recorded and dry biomass was determined by placing the shoots in oven at 80°C for 24 hours. Maximum root length, fibrous root numbers, dry root biomass and shoot height were also obtained.

Table 1. Effects of crumb rubber on medium pH, bulk density, porosity and saturated water holding capacity.

Crumb rubber proportion (%)	pH value	Bulk density (g/ml)	Porosity (%)	Saturated water holding capacity (ml/g)
0	7.75±0.11a	1.42±0.19a	41.2±0.6a	0.56±0.22a
6	7.73±0.06a	1.27±0.10b	38.2±0.9b	0.60±0.19a
9	7.83±0.03a	1.17±0.07c	34.7±0.5c	0.58±0.22a
12	7.80±0.02a	1.07±0.18d	33.7±1.1c	0.62±0.22a
15	7.76±0.02a	1.06±0.16e	35.7±0.8bc	0.60±0.67a
18	7.70±0.03a	1.00±0.07e	29.0±0.8d	0.67±0.39a
21	7.71±0.06a	0.93±0.15f	29.9±1.0d	0.64±0.22a

Results are means ± standard deviation. Means in the same column followed by the same letter are not significantly different at $p<0.05$.

Chlorophyll content and photosynthetic parameters: Chlorophyll was extracted with 1:1 acetone-alcohol and determined spectrophotometrically (Gong, 1995). The net photosynthetic rate (*Pn*), stomatal conductance (*Gs*), transpiration rate (*Tr*), intercellular CO_2 concentration (*Ci*) were determined with a portable photosynthesis system (LICOR-6400; Li-Cor, USA) at a light saturation level with PAR 1000 $umol{\cdot}m^{-2}{\cdot}s^{-1}$. All measurements were made at 08:00 and 10:00 h on a sunny day to avoid any effects of photoinhibition.

Statistical analysis: The data obtained were analyzed using statistical package SPSS (SPSS 12.0; Chicago, IL, USA). The treatment means were compared by Duncan's Multiple Range Test (Duncan, 1955).

Results and Discussion

Crumb rubber didn't affect medium pH value significantly (Table 1). All treatments were in the range of 7.70-7.83. Beyond that a significant reduction ($p<0.05$) in bulk density and total porosity was recorded as the incorporation rate of crumb rubber increased. Maximum reduction of 35% in bulk density and 30% in porosity as compared to control was found at 21% and 18% crumb rubber addition respectively. These results are in agreement with the findings of Baker *et al.*, (2001), who found that crumb rubber incorporation in sand-soil rootzones of sports turf resulted in bulk density and total porosity decrease. While crumb rubber slightly increased saturated water holding capacity, an increase of 20% over control was observed at 18% incorporation rate of crumb rubber. Other researchers had also concluded that physical properties of sand-based putting greens may be improved by inorganic or organic amendments through decreasing bulk density and increasing water holding capacity (Ok *et al.*, 2003; Bigelow *et al.*, 2004). Regression analyses showed that bulk density, total porosity and water holding capacity were interrelated with crumb rubber proportion in linearity. Their correlation coefficients were 0.9875^{**}, 0.9262^{**}, 0.8417^{**} respectively (** denotes $p<0.01$).

Crumb rubber addition had no significant ($p>0.05$) influence on germination rate of *L. perenne*. Germination rates of 0%, 6%, 9%, 12%, 15%, 18%, 21% crumb rubber treatments and control were 75.80%, 75.75%, 75.43%, 78.50%, 78.30%, 77.08% and 72.40% respectively. However, an increase of 3.6%, 3.3% and 1.7% as compared to control was recorded at 12%, 15% and 18% incorporation rates of crumb rubber respectively. Regression analysis on germination rate and crumb rubber proportion reached significant level ($p<0.01$, Fig. 1).

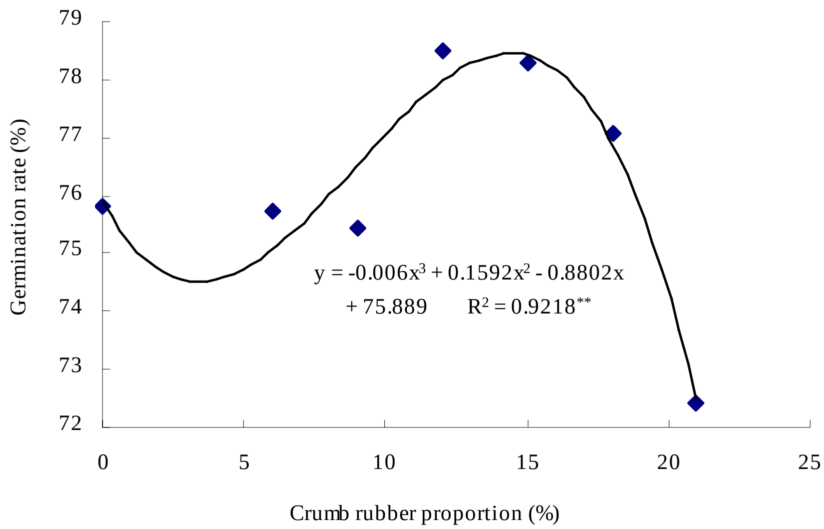

Fig.1. Regressive analysis on germination rate of *Lolium perenne* and crumb rubber proportion (** denotes significant at 0.01 level).

Table 2. Effects of crumb rubber on shoot length and shoot biomass of *Lolium perenne*

Crumb rubber proportion (%)	The first stubble			The second stubble		
	Shoot length (cm)	Fresh weight (g/pot)	Dry weight (g/pot)	Shoot length (cm)	Fresh weight (g/pot)	Dry weight (g/pot)
0	13.62±1.05 a	4.30±0.34 a	0.62±0.06 a	8.83±1.16 a	1.76±0.18 a	0.38±0.04 a
6	12.33±0.85 bc	3.45±0.38 b	0.52±0.06 b	7.89±0.65 b	1.44±0.30 b	0.34±0.03 bc
9	12.20±0.89 bc	3.65±0.13 b	0.57±0.02 ab	7.40±0.98 b	1.30±0.07 bc	0.34±0.01 c
12	12.50±1.10 bc	3.97±0.55 ab	0.56±0.07ab	8.03±1.08 b	1.43±0.06 b	0.35±0.04 abc
15	12.78±1.34 b	4.03±0.38 ab	0.61±0.06 a	7.51±0.86 b	1.47±0.05 b	0.38±0.02 ab
18	11.83±1.25 c	3.71±0.45 b	0.57±0.06 ab	7.54±0.99 b	1.25±0.08 bc	0.33±0.02 c
21	11.95±1.26 c	3.60±0.05 b	0.54±0.04ab	7.32±0.82 b	1.14±0.05 c	0.31±0.01 c

Results are means ± standard deviation. Means in the same column followed by the same letter are not significantly different at $p<0.05$.

A significant reduction ($p<0.05$) in shoot length of the first stubble and the second stubble was evident (Table 2). The first stubble in 15% crumb rubber-treated medium and the second stubble in 12% crumb rubber-treated medium exhibited better growth than other treatments and higher shoot length was obtained, only 6% and 9% lower than their controls respectively. However, at 18% or 21% incorporation rate of crumb rubber, plant growth was retarded remarkably and shoot length reduction up to 14% and 17% as compared to control was recorded for the first stubble and the second stubble respectively.

Table 2 shows the fresh and oven dry biomass of *L. perenne* in the two stubbles as affected by application of different rates of crumb rubber. Crumb rubber incorporation decreased the fresh and oven dry biomass of the two stubbles, especially for the second stubble. At 21% incorporation rate of crumb rubber, maximum reduction of 35% and 20% in fresh weight and oven dry weight as compared to control was observed for the second stubble. However, in all crumb rubber treatments, 15% crumb rubber treatment gave the highest aboveground biomass in two stubbles. A minimum reduction of 6% and 16% in fresh biomass, 4% and 2% in oven dry biomass as compared to control was noted for the first stubble and the second stubble respectively. The adverse effect of crumb rubber on turfgrass growth may be attributed to some toxic substances releasing from crumb rubber. Growth reduction may be generally related with a loss of cellular turgor resulting in either a decrease of mitotic activity or an inhibition of cell elongation (Gabrielli *et al.*, 1990; Shan *et al.*, 2008).

Table 3. Effect of crumb rubber on root growth of *Lolium perenne*.

Crumb rubber proportion (%)	Root length (cm)	Fibrous root number	Root dry weight (g)
0	10.67±1.80 a	6.33±1.23 a	0.82±0.53 b
6	10.88±1.50 a	5.17±1.03 b	0.88±0.15 b
9	10.86±1.51 a	5.50±0.80 ab	1.49±0.64 ab
12	9.41±1.28 ab	6.17±1.59 ab	1.42±0.72 ab
15	10.57±1.23 a	5.58±1.16 ab	1.58±0.12 a
18	10.08±2.19 a	6.17±0.83 ab	1.70±0.14 a
21	8.58±1.92 b	5.58±1.38 ab	1.05±0.09 ab

Results are means ± standard deviation. Means in the same column followed by the same letter are not significantly different at $p<0.05$.

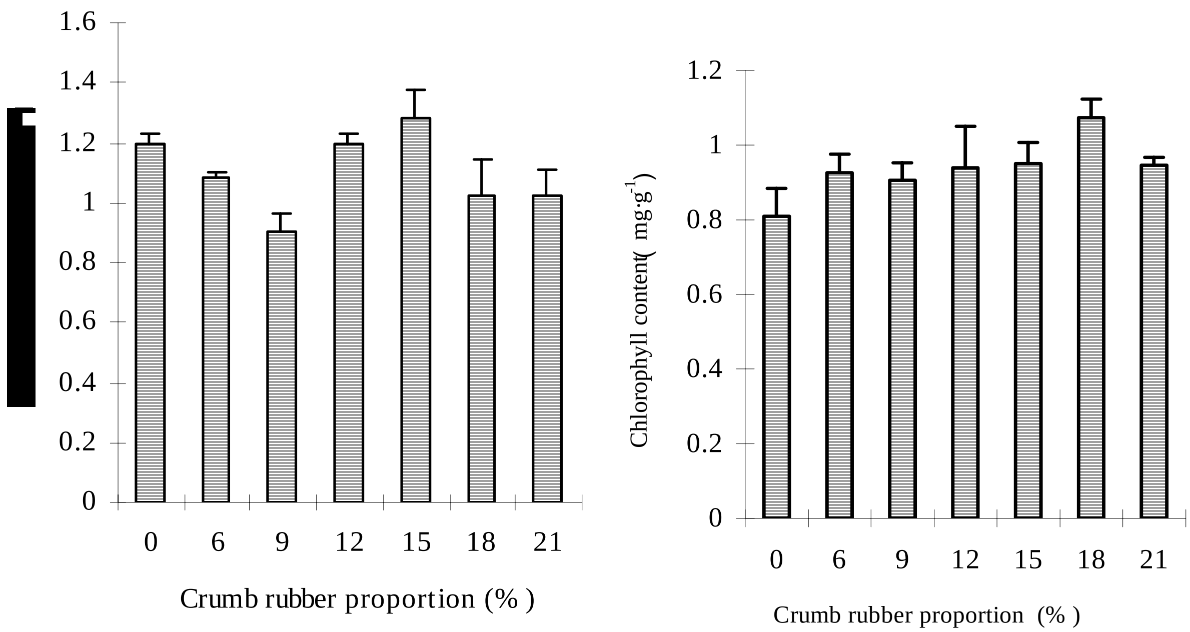

Fig.2. Effects of crumb rubber on chlorophyll content of the first (a) and second (b) stubble of *Lolium perenne*

The second stubble had lower shoot length and biomass than the first stubble. This may be attributed to fertilizer deficiency in the second stubble growth (Table 2). Beard (1973) reported that turfgrass growth must rely on fertilization.

Root length appeared to be reduced beyond 9% crumb rubber, and a significant reduction ($p<0.05$) was found at 21% crumb rubber, with the maximum reduction of 20% over control. Crumb rubber reduced fibrous root numbers of *L. perenne*. Except for 6% crumb rubber treatment, fibrous root number showed no significant difference among other treatments. However, trend of increase in root oven dry weight was observed in all crumb rubber treatments and a significant increase of 94% and 108% over control was recorded at 15% and 18% incorporation rate of crumb rubber. Though crumb rubber inhibited root length and fibrous root numbers, it promoted root weight increase. Li *et al.*, (2000) reported that more root biomass might reserve more nutrients, being beneficial to shoot growth and regeneration. Strong root network formation in topsoil could enhance trampling resistance of turfgrass, and abundant and wide distributed roots could improve drought resistance (Table 3).

Table 4. Effects of crumb rubber on photosynthetic parameters of *Lolium perenne*

Crumb rubber proportion (%)	*Pn* ($\mu mol \cdot m^{-2} \cdot s^{-1}$)	*Gs* ($mol \cdot m^{-2} \cdot s^{-1}$)	*Tr* ($\mu mol \cdot mol^{-1}$)	*Ci* ($\mu mol \cdot mol^{-1}$)
0	6.87±1.21 b	0.082±0.023 b	1.95±0.50 a	287.8±106.7 ab
6	7.07±1.01 b	0.076±0.003 b	1.83±0.06 a	250.3±90.2 bc
9	6.52±0.59 b	0.064±0.005 c	1.50±0.11b	306.7±25.6 a
12	6.50±0.33 b	0.056±0.021 c	2.01±0.18 a	306.8±109.4 a
15	6.99±1.03 b	0.064±0.027 c	1.27±0.47 c	285.5±38.3 ab
18	8.07±1.53 a	0.097±0.009 a	1.26±0.51 c	226.4±73.3 c
21	6.96±0.83 b	0.096±0.034 a	1.99±0.63 a	311.7±79.9 a

Results are means ± standard deviation. Means in the same column followed by the same letter are not significantly different at $p<0.05$.

Chlorophyll content is an important index to evaluate turf quality. It reflects ornamental characteristics and growth status of turf (Li *et al*., 2002). As shown in Fig. 2, an increase in chlorophyll content over control was evident at 12% and 15% incorporation rate of crumb rubber for the first stubble. This was in agreement with the result of seed germination, showing that seed germination may affect chlorophyll synthesis of *L. perenne* in early stage. However, a decline in other crumb rubber treatments can be seen. Maximum reduction of 25% in chlorophyll content over control was found at 9% crumb rubber. Stomatal closure due to elevated CO_2 contents would lead to the chlorophyll reduction (Vitoria *et al.*, 2003). In general, the reduction in chlorophyll contents would be found under stress (Khan *et al.*, 2009). For the second stubble, all crumb rubber treatments enhanced chlorophyll content, especially 18% crumb rubber significantly increased chlorophyll content by 33% as compared to control. With the process of mowing, the synthesis of chlorophyll is promoted, probably because that the negative effect of crumb rubber was weakened and the ability of *L. perenne* adaptation to crumb rubber increased. The increase in photosynthesis of chlorophyll would promote turfgrass shoot growth (Shan *et al*., 2008).

Stoma is a gate for gas exchange of plant leaf and environment (Zhu *et al.*, 2005). The results indicated that *Gs* was characterized by a decrease and then an increase, whereas *Ci* remained high concentration except 18% crumb rubber treatment. *Pn* showed a trend of decrease first and then increase, maximum increase of 17% over control was found at 18% crumb rubber treatment, which was consistent with the result of chlorophyll content in the second stubble. The increase in *Pn* led to the decrease in *Ci* because of the increase in efficiency of CO_2 utilization. However, the decrease in *Pn* was not accompanied by a decrease in *Ci*, (Table 4) indicating that low photosynthetic capacity was due to reduced carboxylation efficiency rather than stomatal limitation (Weng *et al*., 2005).

Conclusions

Lolium perenne L., with high germination rate and strong tillering capacity, can form a very dense, dark green turf rapidly. Mixed with other turfgrasses, *L. perenne* is used extensively for sports field because of its wear resistance and its ability to regenerate. Our results have showed that crumb rubber addition improved medium characters such as bulk density, water holding capacity. Shoot growth and root length were inhibited by the application of crumb rubber, but the effect of 15% crumb rubber was more slightly. While root dry biomass increased in all crumb rubber treatments, the effect of 15% or 18% crumb rubber was more pronounced. More root biomass might be beneficial to shoot growth and regeneration in later stage. Strong root system would enhance the trampling resistance and drought resistance of turfgrass. Maximum chlorophyll content of the first stubble and the second stubble was observed at 15% and 18% crumb rubber treatment respectively. The findings of the current study suggested that the optimum proportion of crumb rubber added in *L. perenne* medium was between 15% and 18%.

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