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Dalbergia sissoo is sensitive to waterlogging as compared to *Bombax ceiba*: A case study under controlled conditions

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Abstract

The total cultivated land area of Pakistan is about 57.99 million hectares (mha). Waterlogging is a major issue that affected almost 16% (more than 1mha) of agriculture soil in Pakistan. Waterlogging is a condition in which the underground water table rises in the root zone or above the soil surface. Therefore, a controlled experiment was conducted to evaluate the tolerance of *Dalbergia sissoo* and *Bombax ceiba* to waterlogging. The growth and biomass production of *Dalbergia sissoo* and *Bombax ceiba* were examined in this study under waterlogging. The results showed that in *Dalbergia sissoo* all the growth (plant height, stem diameter, number of leaves) and biomass production (leaf, stem and root dry weight production) in different plant sections increased significantly under medium water logging (T₁) whereas they decreased significantly under full waterlogged conditions (T₂). In *Bombax ceiba*, all the growth (plant height, stem diameter, number of leaves) and biomass production (leaf, stem and root dry weight production) in different plant sections decreased significantly under medium and full waterlogged conditions. Therefore, it can be concluded that *Bombax ceiba* is more sensitive to waterlogging as compared to *Dalbergia sissoo* and can be preferred in reforestation practices on waterlogged soils.

1. Introduction

Waterlogging is a major issue that affects agricultural productivity throughout the world (Yaduvanshi *et al.*, 2012; Zhang *et al.*, 2016; Jia *et al.*, 2019). It is caused by a variety of factors, including heavy rain, inadequate irrigation, uneven topography, inadequate drainage, and dense texture of the soil. Waterlogging is a condition when the underground water table rises in the root zone or above the soil surface (Pucciariello *et al.*, 2019). In Pakistan, 25% of the irrigated land has been impacted by salinity and waterlogging. 6.17 mha were affected by seasonal flooding or perennial waterlogging, and 1.16 mha of the land have salinity problems in addition to waterlogging issues (Dollinger and Jose, 2018). According to reports, poor irrigation techniques are the major cause of waterlogging in approximately 0.2-0.4% of all irrigated land annually (Arzani, 2008).

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Harmful effects due to waterlogging during the stress event on a broad range of plants' precise features depend on the duration, extent, environmental conditions and level of flooding (Vreugdenhil *et al.*, 2006; Malhotra *et al.*, 2017). Loss of tree foliage in waterlogged areas results in loss of root biomass, leaf necrosis, shedding of leaves, bark damage, as well as imminent susceptibility to insect and fungal pathogens. This ultimately may result in the death of the tree species (Kozłowski, 1997; Parolin, 2001; Kreuzwieser *et al.*, 2004; Parolin and Wittmann, 2010). The extent, nature, and intensity of a flood all affect various plant characteristics during a stressful occurrence (Glick, 2005). Waterlogging harms plant root development and shoot plant growth (Amador *et al.*, 2012; Arbona *et al.*, 2009; Gorjon and Insausti, 2013). One of the main effects is a lack of oxygen or hypoxia caused by waterlogging (Hodge *et al.*, 2009). Excessive flooding leads to a deficiency of O₂ in the rhizosphere for the reason that the flood water in the rhizosphere depletes O₂ rich air from the soil. Plants are exposed to various oxygen levels within the soil during floods and based on how much water is absorbed, they may also suffer anoxia and hypoxia (Parent *et al.*, 2008). Persistent waterlogging interferes with plant respiration rate, hydraulic conductivity, sweating, metabolism of the plant, and photosynthetic rate (Dollinger and Jose, 2018). It also results in the reduction of leaf necrosis, the biomass of roots, leaf drop, damage to the bark, and a significant threat to fungus and insect infections, which results in plant death (Parolin and Wittmann, 2010; Kreuzwieser *et al.*, 2004; Koch *et al.*, 2004).

Different species, though, exhibit tolerance to waterlogging. After some acclimatization, adaptable tree species can continue to produce water and engage in photosynthetic activity (Rasheed *et al.*, 2018). The rate of assimilation becomes reduced during waterlogging stress periods, as noticed in the species of tropical climate and agro temperate ecosystems (Fernandez, 2006; Herrera, 2013). Due to genetic resistance drought drought-tolerant tree species in the flood plains of Amazonia performed physiological functions actively. Such species can coincide with morpho-anatomical variations similar to the manifestation of adventitious roots and hypertrophied lenticels (Kozłowski, 1997; Herrera, 2013). Many tree species that are considered flood-tolerant can develop special hypertrophied lenticels at the bottom of the stem as a reaction to flooding conditions (Glenz *et al.*, 2006).

Trees can ameliorate waterlogged soils through a variety of biological processes (Ferry *et al.*, 2010). The continuation of mitochondrial activity necessitates oxygen intake; this permits the loss of radial oxygen (ROL) from the surface of root cells, which promotes rhizosphere oxidation (Rasheed *et al.*, 2018). Adventitious roots perform crucial tasks, including moisture and nutrient absorption in place of the parent root system's damaged system (Horchani *et al.*, 2008). *Dalbergia sissoo* and *Bombax ceiba* are versatile and economically important tree species that are widely grown. The research will be conducted to assess the possible waterlogging tolerance of species at the sapling stage in terms of physiological and morphological response.

2. Materials and Methods

2.1 Study sites description

The research was conducted from May 2023 to June 2023 in the experimental area of the Department of Forestry and Range Management, University of Agriculture, Faisalabad. Four months old plant saplings were procured from the local nursery. Saplings were left to acclimatize for one week, and fifteen saplings per species (10-12 inches tall) were selected and planted in large containers used in this experiment (Figure 1). Five replicates per species were assigned to either of the three treatments: control (c), Half waterlogged (T1) and full waterlogged (T2). The experiment was conducted under a completely randomized design (CRD) with 3 treatments and 5 replications. In total, there were 30 plants, with 15 plants of each species planted in containers (Figure 1).



Figure 1: Plant of *Dalbergia sissoo* and *Bombax ceiba* in containers.



Figure 2: Growth of *Bombax ceiba* plant in a container

2.2 Application of waterlogging

Large containers with 60 cm diameter and 30 cm depth were used (Figure 1). In control, small holes were made at the base of the container to let the excess water percolate out of the container and to keep the soil at field capacity even under excess watering. Under half waterlogged treatment, small holes were made in the side walls of the container halfway up from the base. This allowed us to keep the soil in the container waterlogged halfway up from the base of the container and allowed the excess water to percolate out of the container halfway down the soil surface. In full waterlogged condition, small holes were made right above the soil surface in the container, which allowed for maintaining a full waterlogging condition just above the soil surface (Figure 1).

2.3 Plant growth and biomass production

Shoot length was calculated for each sapling using a measuring tape from the shoot tip to the root-shoot collar. Due to the relatively small stem thickness, plant diameter was measured for each sampling using a digital caliper at the end of the experiment. After the experiment, the plants were harvested, and their shoot fresh weight was immediately recorded. For this, an electrical weighing balance was used. The average shoot fresh weight for each replication and treatment was then determined. Root fresh weight was measured right away after a gentle washing procedure. The root fresh weight was precisely measured using an electrical weighing balance for each sapling in each treatment. The average root fresh weight for several replications and treatments was calculated. The collected plant shoots were put into paper bags and dried in the sun and then dried in an oven at 70 °C for an additional period until they reached a constant weight in the paper bags. The average shoot dry weight for several replications and treatments was then computed. The collected plant roots were put into paper bags and dried in the sun until their weight remained constant. The dried roots were then dried in an oven for an additional period until they reached a constant weight. The average root dry weight for each replication and treatment was then determined. Root length (cm) was also measured at the end of the experiment. After the soil was washed from the roots, the length of the roots from tip to collar of the plant was measured using a measuring tape. The average root length for several replications and treatments was then determined.

2.4 Statistical analysis

The data of each was analyzed using one-way ANOVA for species effect (Statistica 12.1, Maison Alford, France). Means were compared using Tukey's HSD test and all tests were taken as significant at $P < 0.05$ and were expressed with their standard error.

3. Results

3.1 Plant height

Dalbergia sissoo plants reached a height of 32.6 cm under control conditions, 34.6 cm under T1 (half waterlogged treatment), and 30.6 cm under T2 (full waterlogged treatment). Meanwhile, *Bombax ceiba* plants grew to a height of 34 cm under control conditions, 45 cm under T1, and 31 cm under T2 (Table 1). The results show that both species responded differently to waterlogging treatments. In *Dalbergia sissoo*, the plant height remained similar to the control in T1 but decreased significantly under full waterlogged conditions (T2). In *Bombax ceiba*, a significant increase was evidenced in plant height under half waterlogged conditions (T1) and a significant decrease was evidenced in full waterlogged conditions (T2) compared to the control condition (Table 1).

3.2 Stem Diameter

The results indicated that both species responded differently to stem diameter under waterlogging. In *Dalbergia sissoo*, stem diameter increased significantly to 5.36 mm under T1 (full waterlogged treatment) and remained insignificant at 3.80 mm under T2 (full

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waterlogged treatment). In contrast, *Bombax ceiba* stem diameter remained similar to control under T1 (6.93 mm) and under T2 (6.38 mm; Figure 2A)

3.3 Number of leaves

No significant variation was observed in several leaves in both species. Under controlled conditions, *Dalbergia sissoo* had 99 leaves, which increased to 120 leaves under T1 (half waterlogged treatment) and reached 107 leaves under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had 54 leaves under controlled conditions, slightly increased to 55 leaves under T1 (half waterlogged treatment), and decreased to 43.4 leaves under T2 (full waterlogged treatment; Table 1).

3.4 Leaf fresh and dry weight

The results showed that leaf fresh weight varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a leaf fresh weight of 8.6 grams, which increased to 12.0 grams under T1 (half waterlogged treatment) and decreased significantly to 6.6 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a leaf fresh weight of 36.0 grams under controlled conditions, which significantly decreased to 32.3 grams under T1 and further decreased significantly to 23.12 grams under T2 (full waterlogged treatment, Table 1).

Similarly, leaf dry weight also varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a leaf dry weight of approximately 4.78 grams, which increased significantly to 6.67 grams under T1 (half waterlogged treatment) and decreased significantly to 3.67 grams under T2 (full waterlogged treatment).

3.5 Stem fresh and dry weight

The results showed that stem fresh weight varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a stem fresh weight of 3.60 grams, which increased significantly to 9.00 grams under T1 (half waterlogged treatment) and decreased significantly to 5.50 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a fresh stem weight of 42.0 grams under controlled conditions that decreased significantly to 40.00 grams under T1 and further to 31.00 grams under T2 (full waterlogged treatment; Table 1).

The results showed that stem dry weight varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a stem dry weight of 2.40 grams, which increased significantly to 6.00 grams under T1 (half waterlogged treatment) and decreased significantly to 3.67 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a stem dry weight of 28.00 grams under controlled conditions that decreased significantly to 26.67 grams under T1 and further decreased significantly to 20.67 grams under T2 (full waterlogged treatment Table 1).

Table 1: Growth parameter

	<i>Dalbergia sissoo</i>			<i>Bombax ceiba</i>		
	C	T1	T2	C	T1	T2
Plant height (cm)	32.6 (2.35) a	34 (4.46) a	30.6 (2.01) b	34 (2.23) b	45 (1.14) a	31 (1.43) c
Number of leaves	99 (9.25) c	120 (12.8) a	107 (18.5) b	54 (3.39) a	55 (6.25) a	43.4 (6.96) b
Leaf fresh weight (g)	8.6 (1.07) b	12 (1.41) a	6.6 (1.07) c	36 (1.42) a	32.3 (1.30) b	23.1 (1.01) c
Leaf dry weight (g)	4.77 (0.59) b	6.66 (0.78) a	3.66 (0.59) c	18.4 (0.72) a	16.5 (0.67) b	11.8 (0.52) c
Stem fresh weight (g)	3.6 (0.14) b	9 (1.06) a	5.5 (0.17) c	42 (1.41) a	40 (0.95) b	31 (0.99) c
Stem dry weight (g)	2.4 (0.09) b	6 (0.70) a	3.66 (0.11) c	28 (0.94) a	26.6 (0.94) b	20.6 (0.87) c
Root fresh weight (g)	3.7 (0.10) c	11 (0.35) a	5.7 (0.10) b	43 (2.47) a	34.5 (1.23) b	31 (1.41) c
Root dry weight (g)	2.05 (0.05) c	6.11 (0.19) a	3.16 (0.05) b	18.6 (1.07)	15 (0.53)	13.4 (0.61)

3.6 Root fresh and dry weight

The results showed that fresh root weight varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a fresh root weight of 3.70 grams, which increased significantly to 11.00 grams under T1 (half waterlogged treatment) and decreased significantly to 5.70 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a fresh root weight of 43.00 grams under controlled conditions that decreased significantly to 34.50 grams under T1 and remained at 31.00 grams under T2 (full waterlogged treatment Table 1).

The results showed that root dry weight varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a dry root weight of 2.06 grams, which increased significantly to 6.11 grams under T1 (half waterlogged treatment) and decreased significantly to 3.17 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a dry root weight of

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18.70 grams under controlled conditions that decreased significantly to 15.00 grams under T1, and further decreased to 13.48 grams under T2 (full waterlogged treatment Table 1).

3.7 Total biomass production

The results showed that total biomass production varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a total biomass of 9.23 grams, which increased significantly to 18.78 grams under T1 (half waterlogged treatment) and remained non-significant at 10.50 grams under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a total biomass of approximately 65.16 grams under controlled conditions, which decreased significantly to 58.23 grams under T1, and further decreased significantly to 46.00 grams under T2 (full waterlogged treatment; Figure 2B).

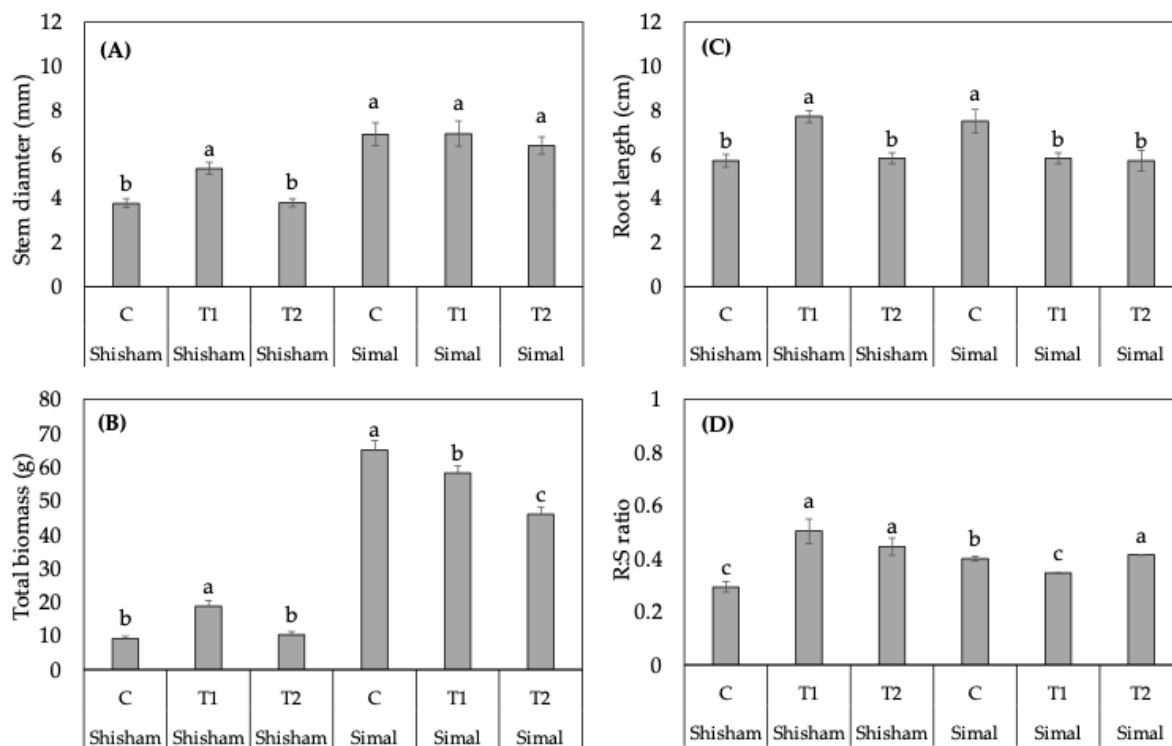


Figure 2: Effect of waterlogging durations on (A) stem diameter, (B) total biomass, (C) Root length and (D) R:S ratio *Dalbergia sissoo* and *Bombax ceiba*.

3.8 Root length

The results showed that root length varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a root length of 14.25 cm, which increased significantly to 19.25 cm under T1 (half waterlogged treatment) and remained non-significantly at 14.5 cm under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a root length of 18.75 cm under controlled conditions, decreasing significantly to 14.5 cm under T1, and remaining at 14.25 cm under T2 (full waterlogged treatment; Figure 2C).

3.9 Root: shoot ratio

The results showed that the root-shoot ratio varied significantly across both species and treatments. Under controlled conditions, *Dalbergia sissoo* had a root-to-shoot ratio of 0.29, which increased significantly to 0.50 under T1 (half waterlogged treatment) and reached 0.44 under T2 (full waterlogged treatment). In contrast, *Bombax ceiba* had a root-to-shoot ratio of approximately 0.40 under controlled conditions, which decreased significantly to 0.35 under T1, and increased non-significantly to 0.41 under T2 (full waterlogged treatment; Figure 2D).

4. Discussion

Waterlogging refers to situations when the water table rises to the root zone of ever above the soil surface (Pucciariello *et al.*, 2019). Furthermore, if the groundwater level is below the depth of the root zone, the capillary zone can spread to the root surface and prevent air circulation due to blockage of soil pores (Rasheed *et al.*, 2018). Waterlogging situation emerges due to many factors. For example, flood irrigation, the absence of concrete-lined canals, and an inadequate drainage system are the main contributing factors to the problem of waterlogging and eventually salinity in the area. There are various forms of waterlogging, such as fluvial flood waterlogging, marine flood waterlogging, seasonal waterlogging, continuous waterlogging, and subterranean waterlogging. During the rainy season, excessive river outflow causes the surrounding lands to waterlog. Similarly, ocean water seeps in the surrounding patches of land, causing waterlogging and sometimes, run off water gathers in the lowlands and depressions during the rainy season to cause seasonal waterlogging.

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The degree of tolerance of waterlogging is different for different species, tree age, waterlogging duration and mobility of water in the soil. Waterlogging results in low oxygen availability to the root system of plants, as roots play an important role in the tolerance of waterlogging. Mitochondrial respiration is responsible for the energy production in waterlogging conditions (Kreuzwieser *et al.*, 2004). In waterlogging conditions, the availability of oxygen is very low during the process of mitochondrial cytochrome c oxidase, probably in between 1-10 10 μ M (Krab *et al.*, 2011; Kreuzwieser *et al.*, 2004). Intolerant of flooding species can gather all the necessary photosynthetic products in various parts, such as leaves, bark and needles rather than transporting to their roots (Sudachkova *et al.*, 2009; Repo *et al.*, 2016a).

Many studies have evidenced the effects of waterlogging on plant growth. In this study, almost all the growth parameters decreased significantly in *Bombax ceiba* species under increased waterlogging. Whereas in *Dalbergia sissoo*, the half-waterlogged condition had a positive impact on plant growth, whereas full waterlogged condition had a significant negative impact of all growth parameters. In the previous studies on three-year-old plants of *Azadiractha excelsa*, a similar negative impact was found on growth under waterlogged conditions however, physiological traits remained unaffected (Shukor, 2014). Similarly, Shaheen *et al.* (2021) evidenced waterlogging tolerance in *Bombax cieba*, *Dalbergia sisso*, *Eucalyptus camaldulensis*, and *Populous deltoides*. The results showed that *Eucalyptus camaldulensis* had the highest potential to withstand waterlogging and revealed that *Bombax cieba*, is very sensitive to waterlogging.

Studies have shown that waterlogging leads to the depletion of oxygen in the root system. Roots represent a primary component of plants, playing a major role in enhancing tolerance to waterlogging in trees. Under water saturation conditions, the maximal energy production in root cells can be substituted by mitochondrial respiration through the maximal fermentation of ethanol (Kreuzwieser *et al.*, 2004). Species vulnerable to flooding accumulate products of maximal photosynthesis in their needles and leaves, which are then transported to the roots (Sudachkova *et al.*, 2009; Repo *et al.*, 2016). Geng *et al.* (2021) worked on assessing the resistance to waterlogging in fifteen poplar clones raised in China's Jiangnan Plain. morphological, development, biomass production, leaf transpiration, and chlorophyll content indices were evaluated under waterlogging. Results revealed that waterlogging treatment drastically altered the morphology of the seedlings, causing the growth of root hairs and lenticels that are enlarged, as well as chlorosis and abscission of leaves. Waterlogging stress also significantly reduced the height, biomass production, and leaf gas exchange parameters, which were in line with the findings of the present study. Studies have shown that waterlogging causes root hypoxia causing serious negative impacts on growth, development, and survival in plants due to O₂-limiting situations. The metabolic conversion from anaerobic to aerobic enzymatic hydrolysis is one of the most well-documented plant responses to soil waterlogging (El-yazel *et al.* 2022). Due to the decreased oxygen availability in root cells caused by waterlogging, plants' development, growth, and production are negatively impacted (Horsham *et al.*, 2009). It leads to a slowdown in photosynthetic activity, stomatal closure (Colmer and Voesenek, 2009), and alterations in other biochemical attributes and cell wall loosening (Komatsu *et al.*, 2011; Palachai *et al.*, 2019). In the current study, it was discovered that for all tree species, normal soils had greater root and shoot length, number of leaves, stem diameter, plant height, and shoot and root weight than saturated soil. About morphological criteria, similar outcomes were reported by Komatsu *et al.* (2011), Horchani *et al.* (2009), and Ashraf and Arfan (2005). These findings align with the results explained by Yordanova *et al.* (2005), who observed that plants exhibited poor performance under waterlogged conditions, leading to stunted growth and development.

Conclusion

The study result revealed that different tree species and duration of waterlogging showed significant variation for growth, physiological and biochemical attributes of various tree species at the sapling stage. The duration of waterlogging has a significant effect on the growth of selected tree species. The longer the duration of waterlogging, maximum was the effect on the plant development, bodily functions, and biochemical properties of various tree species at the sapling stage. Waterlogging duration has the maximum effect on the growth and physiological attributes of the selected tree species. *Dalbergia sissoo* tends to maintain or slightly increase growth and biomass allocation under moderate waterlogged conditions, but severe waterlogging negatively impacts growth. *Bombax ceiba*, on the other hand, shows a decrease in growth and biomass allocation under both moderate and severe waterlogged conditions. These findings contribute to our understanding of how waterlogging affects the growth and physiology of *Dalbergia sissoo* and *Bombax ceiba* plants, which can have implications for environmental management and conservation efforts in waterlogged habitats. Further studies may be needed to explore the underlying physiological and biochemical mechanisms driving these responses.

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