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Atrazine and terbuthylazine mineralization by an *Arthrobacter* sp. isolated from a sugarcane-cultivated soil in Kenya

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ABSTRACT

A tropical soil from a Kenyan sugarcane-cultivated field showed a very high capability to mineralize ¹⁴C-ring-labeled atrazine. In laboratory experiments this soil mineralized about 90% of the applied atrazine within 98 d. The atrazine-degrading microbial community was enriched in liquid cultures containing atrazine as the sole N source and 100 mg L⁻¹ glucose as additional C source. From the enrichment culture a bacterial strain was isolated and identified by comparative sequence analysis of the 16S-rDNA as member of the genus *Arthrobacter*. The enriched mixed culture as well as the isolated strain, designated as *Arthrobacter* sp. strain GZK-1, could grow on atrazine and terbuthylazine as sole N-sources; *Arthrobacter* sp. GZK-1 mineralized ¹⁴C-ring-labeled atrazine up to 88% to ¹⁴CO₂ and ¹⁴C-ring-labeled terbuthylazine up to 65% to ¹⁴CO₂ in a liquid culture within 14 d. The enriched microbial consortium as well as the isolated strain could be a potential solution for the remediation of s-triazine polluted agricultural soils.

1. Introduction

Atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine) has been the most widely used herbicide over the last 50 years. It has been found in groundwater at concentrations exceeding the ground and drinking water limit of 0.1 μ g L⁻¹ of the European Union (Spliid and Koppen, 1998; Close and Rosen, 2001). As a result, the application of atrazine has been forbidden in many European countries. However in Kenya atrazine is still in use since more than 30 years, for example it is applied in sugarcane-cultivated fields situated within major Kenyan river basins.

In the first decades after its introduction atrazine was found to be slowly biodegradable. However, over the past 10 years accelerated atrazine degradation has been observed in fields with a long atrazine history (Barriuso and Houot, 1996). Repeated application of atrazine can thus result in adaptation of the soil microbial community able to rapidly metabolize the compound.

Although atrazine can undergo chemical dechlorination in acidic soils (Wolfe et al., 1990), degradation of atrazine occurs predominantly by biological processes, especially in neutral or slightly alkaline soils. An initial step is the hydrolytic cleavage of the chlorine atom, followed by the sequential removal of the two alkylamino chains resulting in the intermediate product cyanuric acid. The cleavage of the s-triazine ring is a typical biotic reaction;

the microorganisms degrade cyanuric acid stepwise via biuret and allophonic acid into the final end products carbon dioxide and ammonia (Yanze-Kontchou and Gschwind, 1994; Mandelbaum et al., 1995; Radosevich et al., 1995; Cheng et al., 2005; Shapir et al., 2007; Lin et al., 2008). The enzymes and encoding genes involved in s-triazine degradation have been identified (reviewed by Shapir et al., 2007). The genes *trzN/atzA*, *atzB* and *atzC* encode for three hydrolases that catalyze the degradation of s-triazines to cyanuric acid. The enzymes TrzD/AtzD, AtzE and AtzF are necessary for ring cleavage and the hydrolysis of cyanuric acid to CO₂ and NH₃.

Numerous bacteria and a few fungi were isolated from geographically diverse locations which have the capability to partially degrade atrazine and other s-triazines, among them *Pseudomonas* sp. (Mandelbaum et al., 1995), *Nocardioides* sp. (Yamazaki et al., 2008), *Pseudaminobacter* sp. (Topp et al., 2000a), *Clavibacter* sp. (De Souza et al., 1998), *Chelatobacter* sp., *Aminobacter* sp., *Stenotrophomonas* sp. (Rousseaux et al., 2001), *Arthrobacter* sp. (Vaishampayan et al., 2007), *Agrobacterium* sp. (Struthers et al., 1998), *Pleurotus pulmonaris* (Masaphy et al., 1993) and *Phanerochaete chrysosporium* (Mougin et al., 1994). Complete mineralization of atrazine has been reported for two *Pseudomonas* sp. (Yanze-Kontchou and Gschwind, 1994; Mandelbaum et al., 1995) and a bacterial isolate from a pesticide mixing area (Radosevich et al., 1995).

Most of the isolated atrazine-degrading microbes are from temperate soils. There are not much data reported on atrazine-degrading strains from tropical soils (Vaishampayan et al., 2007).

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In Kenya, where atrazine is still widely used, up to now no atrazine-degrading microbial communities or single strains were characterized or isolated. The objective of this study was therefore to investigate the atrazine mineralization capacity of a soil from a sugar cane field with a long atrazine history, to enrich the degrading microbial community from this soil and to isolate and identify the key degrader. The enriched community and the isolated strain should then be tested for its ability to mineralize other s-triazines, namely terbuthylazine (2-chloro-4-ethylamino-6-t-butylamino-striazine), which was never applied on this soil. The atrazine- and terbuthylazine-mineralization capacities of the enriched community as well as the isolated strain should be compared. From a former experiment (Grundmann et al., 2007) it is known that the application of an isoproturon mineralizing microbial community in form of microbial hot spots to various soils was very effective and showed sustainable herbicide mineralization. The data from the present study should show whether the atrazine mineralizing microbial community will have a comparable effective remediation potential as the isoproturon degrading community and the obtained data will also give informations on the applicability of the enriched community and the isolated strain in s-triazine soil remediation which is a subject of great interest.

2. Materials and methods

2.1. Soil

Soil samples were collected in December 2006 from a sugarcane field in Kenya (0–20 cm) where atrazine is used for the last 20 years. The field is situated at 34°50′49″E–35°35′41″E longitude and 0°4′55″N–0°20′11″S latitude and the soil is classified as cambisol with the following characteristics: pH 5.6; 0.14% total N; 1.0% organic C, 76% sand, 14% clay and 10% silt. pH was measured in a water soil suspension and total N and organic C were determined with a C/N analyzer.

Normally, in our laboratory, degradation experiments are conducted at optimum conditions with a soil density of 1.3 g cm⁻³ and a soil water tension of – 15 kPa to ensure maximal pesticide mineralization (Schroll et al., 2006). But the Kenyan soil had a very low solid material density of 2.28 g cm⁻³ as measured with a pycnometer (density bottle). Therefore, this tropical soil could not be compacted to a soil density of 1.3 g cm⁻³ as it is common for Middle European soils. Thus, the Kenyan soil was compacted to a soil density of 1.0 g cm⁻³, which is the typical soil density of this soil in the field, to determine the soil water retention curve (Schroll et al., 2006) and to conduct the degradation experiments.

2.2. Chemicals

Uniformly ^{14}C -ring labeled atrazine (specific radioactivity 6.0×10^8 Bq mmol $^{-1}$; purity >98%) was purchased from Sigma Aldrich (St. Louis Missouri, USA). Uniformly ^{14}C -ring labeled terbuthylazine (specific radioactivity 1.1×10^9 MBq mmol $^{-1}$, purity > 98%) was obtained from Institute of Isotopes, Budapest, Hungary. ^{14}C -atrazine and ^{14}C -terbuthylazine were mixed with the respective non-labeled compounds and dissolved in methanol to obtain appropriate application standards for yielding the final concentrations in the different experiments: 25 mg atrazine kg $^{-1}$ soil, 22 mg atrazine L $^{-1}$ liquid culture and 8 mg terbuthylazine L $^{-1}$ liquid culture. Analytical standards of atrazine and terbuthylazine were purchased from Dr. Ehrenstorfer (Augsburg, Germany). Scintillation cocktails were obtained from Packard (Dreieich, Germany). All other chemicals and solvents were of analytical grade and were purchased from Merck (Darmstadt, Germany).

2.3. Growth media and cell counting

Microbial enrichment steps, isolation procedures and degradation experiments were conducted in basal salts medium (BSM) as previously described by Radosevich et al. (1995) with slight modifications. The medium contained 0.5 g K₂HPO₄, 0.5 g MgSO₄·7H₂O, 15 mg FeCl₃·6H₂O, 11.4 mg CaCl₂·2H₂O, 0.1 mg MnCl₂, and 0.01 mg ZnSO₄·7H₂O per liter of demineralized water.

For cell counting serial dilutions of the liquid culture were spread on Luna Bertani (LB) agar plates and incubated at 30 °C. After 48 h the colony forming units (CFU) were determined. Cells were counted daily in all liquid culture mineralization experiments.

2.4. Mineralization experiments in soil

Mineralization of 14 C-labeled atrazine in soil was studied in a closed laboratory system as previously described by Schroll et al. (2004). Four replicates of soil samples (50 g, dry weight) were incubated in 100 mL double-wall flasks in the dark at 20 ± 1 °C for 98 d. Humidified air ($1.0 \, L \, h^{-1}$) was pumped through the system three times per week for 1 h. After passing through the flasks, the air was conducted through two subsequent absorption tubes which were filled with 0.1 M NaOH solutions to trap 14 CO $_2$ from mineralization process.

For soil incubation experiments methanolic ¹⁴C-atrazine-solution was applied drop-wise onto 3.5 g of previously dried (105 °C, 24 h) ground soil to give a final atrazine concentration of $25 \text{ mg kg}^{-1} \text{ soil } (21 \times 10^5 \text{ Bq kg}^{-1})$. After evaporation of methanol the soil sample was mixed intensively. The soil aliquot was then transferred to 46.5 g (dry weight) pre-equilibrated soil (20 ± 1 °C, -35 kPa), well mixed with a spatula and filled into incubation flasks. Then, soils were compacted with a spatula to a volume equivalent to a soil density of 1.0 g cm⁻³ and adjusted to a water tension of -15 kPa. Soil moisture was controlled gravimetrically during the whole experiment. At the end of experiments, extractable and non-extractable ¹⁴C-pesticide residues were quantified. From each replicate an aliquot of 35 g soil material (dry weight) was extracted by accelerated solvent extraction (ASE 200, Dionex, Sunnyvale, USA) and the radioactivity in the extracts was measured by liquid scintillation counting (Tricarb 1900 TR, Packard, Dreieich, Germany). Aliquots of the extracted soils were air-dried and combusted (Oxidizer 306, Packard, Dreieich, Germany) to quantify the amount of non-extractable pesticide residues.

2.5. Enrichment of the degrading microbial community, isolation of strains and mineralization experiments with the isolates

The bacterial community from the Kenyan soil was enriched in basal salts medium (BSM) with ¹⁴C-atrazine as the sole nitrogen source and 100 mg L^{-1} glucose as carbon source. 55 μL of methanolic ¹⁴C-atrazine-solution (final concentration in liquid culture: 22 mg L^{-1} , $6.8 \times 10^4 \text{ Bq L}^{-1}$) was applied in a sterile 100 mL-Erlenmeyer flask and methanol was left to evaporate. The resultant atrazine crystals were dissolved in 25 mL sterile BSM by shaking on an orbital shaker (GFL, Burgwedel, Germany) over night. Thereafter, two aliquots of 100 µL BSM were taken for radioactivity measurement to determine the initial ¹⁴C-atrazine concentration. Subsequently, 5 g of soil (dry weight; equilibrated for 10 d at 20 °C and -15 kPa) were added to the medium and incubated on an orbital shaker at 100 rpm in the dark at 20 ± 1 °C. The incubation flasks were connected daily to the closed laboratory system, as described in Section 2.4, and aerated for 1 h to trap the evolved ¹⁴CO₂ and to report the atrazine mineralization ability of the bacterial community. Sterile filters (0.22 µm, Sartorius, Göttingen, Germany) were connected at the air inlet and air outlet of the incubation flasks

to ensure sterile conditions in the liquid cultures. The experiments were conducted in quadruplicates. After incubating the soil suspension for 30 d, 1 mL was transferred to 24 mL of fresh growth medium; the following liquid cultures were transferred weekly to fresh media.

From the 10th enrichment culture 1 mL aliquots were taken to inoculate fresh, sterile growth media (24 mL) which were spiked with ^{14}C -terbuthylazine (8 mg L $^{-1}$, 6.8 \times 10 4 Bq L $^{-1}$) and mineralization experiments were conducted with daily sampling of $^{14}\text{CO}_2$ to investigate whether the enriched microbial community was able to mineralize beside atrazine another s-triazine, namely terbuthylazine.

The 10th enrichment culture was also used for isolation of bacterial strains: aliquots were taken and serial dilutions were spread on BSM agar plates (1.5% agar, 22 mg atrazine $\rm L^{-1}$). The plates were incubated in the dark at 30 °C for at least 48 h. Single colonies were transferred using a sterile inoculation loop onto BSM agar plates to end in pure cultures. Obtained strains were re-transferred to liquid cultures and their atrazine and terbuthylazine mineralizing capacity was tested in further mineralization experiments.

The initial concentrations of atrazine ($22~mg~L^{-1}$) and terbuthylazine ($8~mg~L^{-1}$) in the liquid culture mineralization experiments were different, depending on their different water solubility, but the chemical substrate activities were nearly equal (0.8~and~0.9, respectively).

2.6. DNA extraction, PCR amplification and sequencing of 16S-rDNA coding genes

High chromosomal weight DNA was extracted from the isolated strain by using the Fast DNA®SPIN kit for soil (MP Biomedicals, Eschwege, Germany). The universal primer pair 616F, 5'-AGA-GTTTGATYMTGGCTCAG 5' and 630R, 5'-CAKAAAGGAGGTGATCC 3' (Juretschko et al., 1998), corresponding to nucleotide positions 8–27 and 1528–1544 in the 16S-rDNA from *E. coli* (Brosius et al., 1981), was used for amplification of a 1.4 kb region of the 16S-rRNA coding genes.

The amplified 16S-rDNA gene fragments were further gel-purified and subsequently cloned with the TOPO TA Cloning® kit containing pCR®2.1 vector (Invitrogen, Paisley, UK). Insert-positive clones (blue-white selection) were isolated and purified with the Nucleo spin Plasmid miniprep Isolation kit (Macherey-Nagel, Dueren, Germany). The presence of inserts with correct size was controlled as described in the manufacturer's protocol by *EcoRI* restriction endonuclease digestions. 16S-rDNA plasmid inserts were sequenced using the Big-Dye-Terminator sequencing Kit (Applied Biosystems, Darmstadt, Germany) and an ABI-Prism-377 automated Sequencer (Applied Biosystems, Darmstadt, Germany) as it was previously described (Sharma et al., 2008).

The obtained sequence was further analyzed phylogenetically using the software package ARB (http://www.arb-home.de) (Ludwig et al., 2004). Sequence was added to an existing database of well aligned small-subunit rRNA gene sequences (SILVA_95) (Pruesse et al., 2007) by using the fast alignment tool implemented in the ARB software package.

3. Results and discussion

3.1. Atrazine mineralization in soil

The soil had a high capability for mineralizing atrazine. In laboratory experiments about 90% of the applied ¹⁴C-ring-labeled atrazine was mineralized after 98 d of incubation. The remaining soil residues showed 2.6% extractable radioactivity and 7.4% non-extractable radioactivity. The indigenous microbial community

was obviously able to degrade atrazine very efficiently after 20 years atrazine history. Thus, our findings confirm former results, which showed that in soils repeatedly treated with atrazine, adapted degrading microorganisms are established, resulting in an enhanced atrazine degradation (Barriuso and Houot, 1996).

3.2. Isolation and characterization of an atrazine and terbuthylazine mineralizing strain

The isolation of bacterial strains was started from the 10th enrichment culture. Twelve strains were isolated on BSM agar plates which were re-transferred to liquid culture to test their atrazine mineralizing capacities. Only strain GZK-1 was able to mineralize ¹⁴C-ring-labeled atrazine. In additional experiments it could be shown that the isolated strain was also able to mineralize ¹⁴Cring-labeled terbuthylazine. It could grow on atrazine and terbuthylazine as the sole nitrogen sources. The isolated strain GZK-1 was characterized by comparative sequence analysis of the 16SrRNA coding genes as member of the genus Arthrobacter. Strain Arthrobacter sp. GZK-1 (accession number of the 16S-rRNA sequence FJ766438) shows the highest similarity of 99.2% to a sequence derived from an uncultured Actinobacterium (accession number AY622266) and exhibit 99.0% similarity to the next related cultured bacterium, Arthrobacter sp. SMCC G964 (accession number AF197029).

Arthrobacter strains were frequently isolated from soil and are commonly found at sites polluted with chemicals and radioactive materials. They are known to survive under environmentally harsh conditions. Mongodin et al. (2006) described the genome of Arthrobacter aurescens strain TC1 and they found that the genome of this bacterium consists of a single circular chromosome and two plasmids that harbour a large number of genes coding proteins that are involved in stress responses due to starvation, desiccation, oxygen radicals and toxic chemicals. Arthrobacter sp. are further able to degrade a variety of environmental pollutants such as methyl t-butyl ether, phthalate, nitroglycerin and a large number of s-triazine herbicides (Eaton, 2001; Liu et al., 2001; Marshall and White, 2001; Strong et al., 2002).

Several Arthrobacter sp. were isolated from agricultural soils, rhizosphere, spill sites and wastewater in France, Canada, USA, China, New Zealand, and India which were able to start the degradation process but could not totally degrade ¹⁴C-ring-labeled atrazine to ¹⁴CO₂ and NH₃: Arthrobacter crystallopoietes (Rousseaux et al., 2001), Arthrobacter aurescens TC1 (Strong et al., 2002), Arthrobacter sp. AD1 (Cai et al., 2003), Arthrobacter nicotinovorans HIM (Aislabie et al., 2005), Arthrobacter sp. strain MCM B-436 (Vaishampayan et al., 2007). Those species carry the s-triazinedegrading genes atzA, atzBC or atzABC which enable them to degrade atrazine to cyanuric acid. No Arthrobacter sp. was reported to hydrolyze cyanuric acid and to yield carbon dioxide and ammonia. The Arthrobacter sp. GZK-1 isolated in this work was able to mineralize atrazine, but the enzymatic arsenal of this strain cannot be described, since up to now the degrading genes were not identified.

Moreover, the isolated *Arthrobacter* sp. GZK-1 could also mineralize terbuthylazine. Previous studies (Dousset et al., 1997; Gerstl et al., 1997; Langenbach et al., 2001) found terbuthylazine to be poorly mineralized even in soils which had been exposed to the herbicide. Among the isolated atrazine degraders some species, like *Pseudaminobacter* sp. (Topp et al., 2000a), *Nocardioides* sp. (Topp et al., 2000b), *A. nicotinovorans* HIM (Aislabie et al., 2005) and *Arthrobacter aurescens* TC1 (Strong et al., 2002) could degrade terbuthylazine. *Pseudomonas* sp. strain ADP was reported to mineralize terbuthylazine (Cabrera et al., 2008) but to a lower extent than it could mineralize atrazine. *Arthrobacter* sp. strain GZK-1 isolated in this study is thus another strain that has the capability for

complete degradation and mineralization of atrazine and terbuthylazine.

3.3. Atrazine mineralization capacity of the enriched microbial community and the isolated Arthrobacter sp. in liquid culture

The microbial community as well as the isolated *Arthrobacter* strain maintained their atrazine and terbuthylazine mineralization function very stable during many re-inoculation steps over 18 months. The data shown in Figs. 1–3 were obtained after 20 inoculation cycles.

The microbial community reached the maximum mineralization rate of around 380 μ g g⁻¹ within 1 d, whereas the *Arthrobacter* sp. cells showed their maximum degradation performance of 280 $\mu g g^{-1}$ just after 3 d (Fig. 1). Since more Arthrobacter sp. cells were measured in the liquid culture with the single strain than total cells in the liquid culture with the microbial community (Table 1) it could be reasoned that there exist less Arthrobacter sp. cells in the community than in the experiment with the single strain. But nevertheless, the microbial community mineralized atrazine faster than the isolated strain. This is a strong hint that there exists an interaction of several microbial strains present in the community that enhance the mineralization function of the key degrader. In literature multi member consortia, isolated and enriched from an agricultural soil with a long history of atrazine exposition (De Souza et al., 1998; Smith et al., 2005) or from an agrochemical factory soil (Kolic et al., 2007) were investigated to elucidate bacterial community interactions that may be involved in atrazine degradation. All

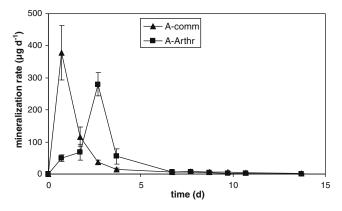


Fig. 1. Mineralization rates of atrazine in liquid culture with the degrading community (A-comm) and with the isolated *Arthrobacter* strain (A-Arthr).

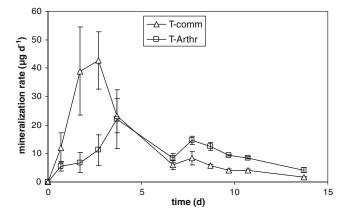
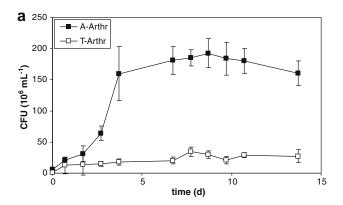


Fig. 2. Mineralization rates of terbuthylazine in liquid culture with the degrading community (T-comm) and with the isolated *Arthrobacter* strain (T-Arthr).



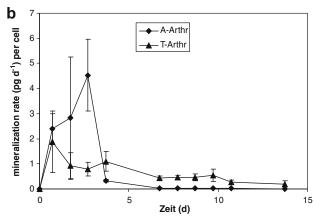


Fig. 3. Comparison of the atrazine and terbuthylazine mineralization capacity of *Arthrobacter* sp. strain GZK-1: (a) number of *Arthrobacter* sp. GZK-1 cells in liquid culture with atrazine (A-Arthr) and in liquid culture with terbuthylazine (T-Arthr); (b) mineralization rate of atrazine (A-Arthr) and terbuthylazine (T-Arthr) in liquid culture per *Arthrobacter* sp. cell.

studies showed that complete atrazine mineralization was a result of the combined metabolic attack of the different members of the consortia on the atrazine molecule and that complex cooperation and interactions exist between the community members. De Souza et al. (1998) also found that the consortium metabolized atrazine faster than did a single strain.

The microbial community consumed the atrazine-substrate very quickly and reached thus the highest possible atrazine mineralization rate in a shorter time than the single strain. Nevertheless, the cumulative atrazine mineralization within 14 d was about 88% (Table 1) and nearly identical for both, microbial community and single strain. Mandelbaum et al. (1995) found a similar high mineralization of 80% for *Pseudomonas* sp. ADP, which was isolated from a herbicide spill site, whereas a bacterial isolate from a surface soil from a pesticide mixing aerea (Radosevich et al., 1995) and *Pseudomonas* strain YAYA6 (Yanze-Kontchou and Gschwind (1994) showed a lower atrazine mineralization capacity of 40–50% and about 50%, respectively.

3.4. Terbuthylazine mineralization capacity of the enriched microbial community and the isolated Arthrobacter sp. in liquid culture

The microbial community showed a higher terbuthylazine mineralization potential (79%) within 14 d than the single *Arthrobacter* strain (65%, Table 1). This is mostly due to the different mineralization rates at the beginning of the degradation process. The microbial consortium exhibited significantly higher terbuthylazine mineralization rates in the first 3 d of incubation than the isolated strain (Fig. 2). Thus, the cumulative mineralization in this 3 d time

 Table 1

 Cumulative mineralization of atrazine and terbuthylazine in liquid culture with active microbial community or isolated Arthrobacter sp. strain GZK-1 (incubation time: 14 d).

	Mineralization (% of applied ¹⁴ C)	Cell counts	
		Beginning of experiment (10 ⁶ CFU mL ⁻¹)	End of experiment (10^6 CFU mL $^{-1}$)
Atrazine microbial community	87.7 ± 8.8	2.2 ± 0.9	97.6 ± 21.9
Atrazine Arthrobacter sp.	86.7 ± 9.7	6.0 ± 0.4	160.0 ± 20.0
Terbuthylazine microbial community	79.3 ± 10.1	3.4 ± 1.7	31.1 ± 24.5
Terbuthylazine Arthrobacter sp.	64.6 ± 6.2	0.6 ± 0.3	27.0 ± 10.4

span was much higher for the microbial community (45%) in comparison to the experiment with the *Arthrobacter* sp. strain (12%). Total cell numbers at the beginning and at the end of both experiments did not significantly differ (Table 1) and it can be assumed that the number of *Arthrobacter* sp. GZK-1 cells in the community is most likely lower than the amount of *Arthrobacter* sp. cells in the experiment with the isolated strain, as it was the case for atrazine. Similar results from the liquid culture experiments on atrazine and terbuthylazine mineralization capacity of the microbial consortium and the single strain allows to conclude that the mineralization of both s-triazines is more effective when the microbial community is applied.

3.5. Capacity of Arthrobacter sp. strain GZK-1 for atrazine and terbuthylazine mineralization

Arthrobacter sp. strain GZK-1 could grow on atrazine much better than on terbuthylazine (Fig. 3a). In liquid cultures with terbuthylazine as the sole N-source the cell number increased slowly and leveled at a cell count of 27×10^6 CFU mL $^{-1}$, whereas in liquid cultures with atrazine as the sole N-source the cells reached relatively fast a higher value of 160×10^6 CFU mL $^{-1}$.

By comparing the mineralization rates of atrazine and terbuthylazine per cell, Fig. 3b shows clearly that *Arthrobacter* sp. GZK-1 can mineralize atrazine more effectively than terbuthylazine. Since the molecules only differ in an alkyl side chain, it seems that the enzyme responsible for the removal of this side chain can more easily remove the isopropyl group from the atrazine molecule than the t-butyl group from the terbuthylazine molecule.

4. Conclusions

Arthrobacter sp. strain GZK-1 can mineralize the s-triazines atrazine and terbuthylazine. Arthrobacter sp. are known for their ability to survive under environmentally harsh conditions and to degrade a variety of environmental pollutants. Several Arthrobacter sp. are reported in literature to degrade s-triazines but they cannot mineralize atrazine or terbuthylazine. Apparently, Arthrobacter sp. strain GZK-1, that was isolated from a Kenyan sugar cane field with a long history of atrazine use, is a phenotype that has acquired the enzymatic arsenal for complete degradation of atrazine and terbuthylazine. His potential for mineralization of other s-triazines has to be tested. So, Arthrobacter sp. GZK-1 is a new candidate for bioremediation of s-triazine contaminated agricultural soils.

The obtained data give some hints that the application of the enriched microbial community might be more efficient in the mineralization of both s-triazines than the isolated strain. Taken together with results from a previous study (Grundmann et al., 2007), it could be hypothesized that the inoculation of agricultural soils with specific pesticide degrading microbial communities might be a very effective and sustainable approach for the enhanced degradation of organic soil contaminants. Thus, inoculation experiments with the isolated *Arthrobacter* sp. strain GZK-1 as well as with the enriched microbial community must be conducted to prove this hypothesis.

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