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Evidence for dose-dependent effects on plant growth by *Stenotrophomonas* strains from different origins

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ABSTRACT

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Aims: To assess the influence of *Stenotrophomonas* on plants, the interaction of 16 *Stenotrophomonas* strains from clinical and environmental sources with strawberry plant seedlings was analysed.

Methods and Results: *In vitro*, all *Stenotrophomonas* strains influenced plant growth when applied to seedlings. Whereas most of the *Stenotrophomonas* strains promoted root growth and hair development, a statistically significantly negative influence on the length of stem was found. Although strains from a clinical origin also showed statistically significant effects on plants, this was generally lower when compared with environmental strains. For three selected strains, a strong dose-dependent effect was observed for all parameters. *In vitro*, a correlation was found between plant growth promotion and production of a plant growth hormone, indole-3-acetic acid (IAA). *Xanthomonas campestris*, a phylogenetically very closely related species to *Stenotrophomonas*, was used as a phytopathogenic control. It too confirmed the reduction of plant growth in this *in vitro* system.

Conclusions: Independent of their origin, *Stenotrophomonas* strains can produce IAA *in vitro* and subsequently, influence plant growth. The effect of *Stenotrophomonas* presence on plants was dose-dependent.

Significance and Impact of the Study: The dose-dependent effect of *Stenotrophomonas*, a bacterium of both biotechnological and medical interest, is of great interest for biocontrol applications of plant-associated strains. This paper is the first report that clearly demonstrates the phytopathogenic capacity of *Stenotrophomonas*.

Keywords: indole-3-acetic acid, plant–bacterium interaction, *Stenotrophomonas*, strawberry seedling assay.

INTRODUCTION

Stenotrophomonas species play an important role in nature, especially in the element cycle; however, nowadays, they are also frequently used in applied microbiology and biotechnology (Ikemoto *et al.* 1980). The biotechnological importance of *Stenotrophomonas maltophilia* is partly due to their potential plant growth-promoting effects and their use in biological control of plant fungal diseases (Berg *et al.* 1994; Kobayashi *et al.* 1995; Nakayama *et al.* 1999). Recent interest has also focused on the capability of *Stenotrophomonas* to degrade xenobiotic compounds (Binks *et al.* 1995) and their potential for decontaminating soil (bioremediation). Also, in the last decade, *S. maltophilia* has become increasingly multidrug-resistant, causing significant case/fatality ratios in certain patient populations, particularly in those who are severely debilitated or immunosuppressed (for a review see Denton and Kerr 1998).

The genus *Stenotrophomonas* is phylogenetically placed in the γ-subclass of *Proteobacteria* (Moore *et al.* 1997). The *Stenotrophomonas* genus was described with the species *S. maltophilia* (Palleroni and Bradbury 1993), previously called *Pseudomonas maltophilia* (Hugh and Ryschenko 1961) and later changed to *Xanthomonas maltophilia* (Swings *et al.* 1983). New *Stenotrophomonas* species were also described by Drancourt *et al.* (1997), Finkmann *et al.* (2000) and Wolf

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et al. (2002). By analysing their 16S rDNA sequence, it was possible to distinguish different genomovars of *S. maltophilia* (Minkwitz and Berg 2001). So far, all validly described *Stenotrophomonas* species have shown a high intraspecific diversity and heterogeneity on the phenotypic and genotypic level (Berg *et al.* 1999; Hauben *et al.* 1999).

In contrast to the extremely closely related genus *Xanthomonas*, no phytopathogenic capacity of *Stenotrophomonas* is known; at present, this organism is not regarded as a plant pathogen (Palleroni and Bradbury 1993). Conversely, all species currently included in the genus *Xanthomonas* are highly specialized plant pathogens (Van den Mooter and Swings 1990; Van Sluys *et al.* 2002). Hence, the absence of phytopathogenic properties of *Stenotrophomonas* was one of the main reasons to exclude the species from *Xanthomonas* to which it had formerly belonged (Van Zyl and Steyn 1992; Palleroni and Bradbury 1993). Although, to date, there is no published evidence that *Stenotrophomonas* strains cause diseases in plants, during a field trail using *Stenotrophomonas* as a plant growth promotion and biocontrol of soilborne pathogens, we observed wilting in strawberry plant after *Stenotrophomonas* treatment on their roots.

In an attempt to clarify this observation, we analysed the interaction of *S. maltophilia* and *S. rhizophila* strains with strawberry plant seedlings using a microplate seedling assay and compared them with the phytopathogenic *X. campestris*. First, the influence of *Stenotrophomonas* application on plant growth was assessed by measuring root and stem length and counting leaves and root hairs. Secondly, three selected strains were investigated in a dose-dependent fashion. Thirdly, the production of the plant growth hormone indole-3-acetic acid (IAA) by bacterial strains was investigated. *Stenotrophomonas* strains from both different plant microenvironments and clinical origins were investigated. The latter group was of particular interest regarding level of interaction and pathogenicity with humans compared with plants; a phenomenon previously found for plant- and human-associated *Burkholderia* and *Pseudomonas* strains (Cao *et al.* 2001). In cases when bacterial interaction between human and plants is mutually exclusive, it is important to identify properties that distinguish between clinical and environmental strains. Here, our goal was to assess the effect of *Stenotrophomonas* on plants and to see whether this depends on the origin of strains and/or concentration applied. The data provided here are of importance for potential uses of *Stenotrophomonas* in biotechnology and in regard to taxonomical questions.

MATERIALS AND METHODS

Isolates and reference strains

A total of 16 clinical and environmental isolates of *Stenotrophomonas* were investigated (Table 1). The clinical strains were isolated in the Rigshospitalet, Copenhagen, Denmark, from various sites [tracheal aspirates, sputa, blood, throat, wounds, skin, ulcers, drainage fluids and aspirates, catheters, urine, etc. (Gerner-Smidt *et al.* 1995)]. *Stenotrophomonas maltophilia* DSM 50170^T (ATCC 13637, a type strain isolated from pleural fluid of a patient with oral carcinoma (Hugh and Ryschenko 1961), *S. nitritireducens* DSM 12575^T [a type strain isolated from an ammonia supplied biofilter (Finkmann *et al.* 2000)], and *X. campestris* DSM 3586^T were used as reference strains.

Table 1 *Stenotrophomonas* strains investigated in this study, their origin, relevant references and clustering into genomovars

Strain	Name	Origin	Reference	Genomovar
e-p14	*S. rhizophila*	Rhizosphere of potato (Lüsewitz 1996)	Minkwitz and Berg (2001)	E1
e-p17	*S. rhizophila*	Geocaulosphere of potato (Lüsewitz 1996)	Minkwitz and Berg (2001)	E1
e-p3	*S. maltophilia*	Rhizosphere of rape (Poel 1989)	Minkwitz and Berg (2001)	E2
e-p19	*S. maltophilia*	Geocaulosphere of rape (Lüsewitz 1996)	Minkwitz and Berg (2001)	E2
e-p20	*S. maltophilia*	Rhizosphere of potato (Braunschweig 1998)	Berg *et al.* (2001)	E2
c5	*S. maltophilia*	Human (Copenhagen 1995)	Gerner-Smidt *et al.* (1995)	C
c6	*S. maltophilia*	Human (Copenhagen 1995)	Gerner-Smidt *et al.* (1995)	C
c20	*S. maltophilia*	Human (Copenhagen 1995)	Gerner-Smidt *et al.* (1995)	C
e-a1	*S. maltophilia*	Brakish water (Zingst 1996)	Minkwitz and Berg (2001)	C
e-a22	*S. maltophilia*	Seawage (Braunschweig 1999)	Minkwitz and Berg (2001)	C
e-a21	*S. maltophilia*	Seawage (Braunschweig 1999)	Minkwitz and Berg (2001)	C
e-a23	*S. maltophilia*	Eye care solution (Munich 1999)	Bader *et al.* (1999)	C
e-p13	*S. maltophilia*	Rhizosphere of rape (Rostock 1995)	Minkwitz and Berg (2001)	C
DSM 50170^T	*S. maltophilia*	Human (1961)	Hugh and Ryschenko (1961)	C
DSM 14405^T	*S. rhizophila*	Rhizosphere of rape (Rostock 1993)	Wolf *et al.* (2002)	E1
DSM 12575^T	*S. nitritireducens*	Ammonia-supplied biofilters	Finkmann *et al.* (2000)	
DSM 3586^T	*Xanthomonas campestris*	*Brassica oleracea*	Skerman *et al.* (1980)	

All *Stenotrophomonas* isolates were identified using the API (BioMérieux, Mercy Etoile, France) and BIOLOG identification systems (Biolog, Inc., Haward, CA, USA) as in Berg *et al.* (1999). 16S rDNA sequence confirmed their taxonomic grouping in *Stenotrophomonas* (Minkwitz and Berg 2001). Unless otherwise stated, isolates were routinely grown in nutrient broth 2 (Sifin, Berlin, Germany) and stored in broth containing 15% glycerol at −70°C.

Phytochamber assay to assess the effect of *Stenotrophomonas* on plant growth

Strawberry cv. Rügen Selecta (Erfurter Samenzucht, Erfurt, Germany) seeds were pregerminated in moist chambers at 25°C for 6 days. Three standard 24-well microplates (Roth, Karlsruhe, Germany) were filled with 1 ml water/agar [20 g agar (Difco), 1 l distilled water, pH 6·8]. To each well, one pregerminated seed followed by 10 μl bacterial suspension (nutrient broth 2, grown for 18 h) was added. *Stenotrophomonas* was evaluated at 10^3, 10^5, 10^7 and 10^9 CFU ml^{-1} and compared with a control using 10 μl of 0·9% NaCl. To determine the effects of bacterial treatment on plant growth, stem and root length was measured and, first, leaves and radial roots were counted (according to Berg *et al.* 2001) 5 weeks after incubation (16 h artificial light, 22°C) in a phytochamber (Percival Scientific, Boone, IA, USA). Each strain was tested at each concentration in 24 replicates and the experiment was repeated in triplicate.

IAA production by *Stenotrophomonas* strains

The microplate method used was a modification of the method developed by Sawar and Kremer (1995). All isolates were grown on half-strength tryptic soya agar (Gibco, Paisley, UK). For the colorimetric IAA assay, a 24-h culture of each isolate was suspended in sterile PBS to an optical density of approx. 0·5 at 500 nm. The suspension (500 μl) was added to 5 ml of growth medium, which contained 5 g glucose, 0·025 g yeast extract, 0·204 g L-tryptophan per 1 l demineralized water. Controls were prepared by substituting bacterial suspension with sterile water. Tubes were incubated in the dark at 20°C for 72 h. Prior to analyse for IAA equivalents, 1 ml growth medium of each isolate was centrifuged at 4°C for 10 min. The supernatant of each sample (150 μl) was dispensed into 96-well microplates followed by the addition of 100 μl of Salkowski reagent (2 ml 0·5 M $FeCl_3$ mixed with 98 ml 35% perchloric acid). Following reaction for 30 min, the colour intensity was measured at 530 nm on a microplate reader (Spectramax-250; Molecular Devices, Sunnyvale, CA, USA). The reproducibility of the results was verified in three independent experiments using one 96-well microplates for each strain.

Statistical analysis

Data on plant growth promotion were analysed for significance using Mann–Whitney *U*-test ($P \leq 0{\cdot}05$) by Statistical Product and Service Solutions for Windows, Rel. 9.0.1. (SPSS, Inc., Chicago, IL, USA). A polynomic regression analysis was used to compare the treatments at each *Stenotrophomonas* concentration in the phytochamber assay.

RESULTS

Effect of *Stenotrophomonas* treatment on plant growth

A total of 16 *Stenotrophomonas* strains from different sources including the four type strains (*S. maltophilia* DSM 50170^T, *S. rhizophila* DSM 14405^T, *S. nitritireducens* DSM 12575^T and the phytopathogenic *X. campestris* DSM 3586^T, Table 1) were characterized by their influence on plant growth. A bioassay to evaluate the plant growth promoting effect by *Stenotrophomonas* strains was used, and strawberry seeds either untreated or treated with *Stenotrophomonas* were germinated in microplates in a phytochamber. In a first screening step, the influence on plant growth was assessed using a bacterial concentration of 10^5 CFU ml^{-1}. In our preliminary studies, this concentration was used as it was found to be optimal to assess rhizobacteria influence on plants (Berg *et al.* 2001). The effect of bacteria on the growth of seedlings was monitored by measuring root and stem length, and by counting leafs and root hairs (Fig. 1a–d). Most isolates (82%) induced increased root length compared with the nontreated control (Fig. 1a). Isolates of the genomovar C, which are of clinical and aquatic origin, exerted the most effect. Of these, the *S. maltophilia* isolate e-a23, originally isolated from clinical eye care solution, enhanced root length by 44% (8·06/5·61 mm nontreated control). The type strain of *S. maltophilia* DSM 50170, which was isolated from human pleural fluid, enhanced the length of the root by 38%. As expected, two strains statistically significantly reduced root length, one of them, phytopathogenic *X. campestris*, reduced the length by as much as 24%. Effects on stem length were mixed, with only a slight majority of *Stenotrophomonas* isolates demonstrating a positive effect and 47% having a negative effect (Fig. 1b). Once again the *Xanthomonas* treatment had the most negative effect and reduced length by 34% (9·12/13·89 mm nontreated control). Additionally, first leaf development was influenced by 65% of *Stenotrophomonas* isolates (Fig. 1c) and root hair development by 71% of the isolates (Fig. 1d), although only 24% showed statistically significant effects. The plant-associated isolates DSM 14405 and e-p17 showed the highest effect on leaf development, i.e. 21 and 12%, respectively. They also had an extremely strong effect

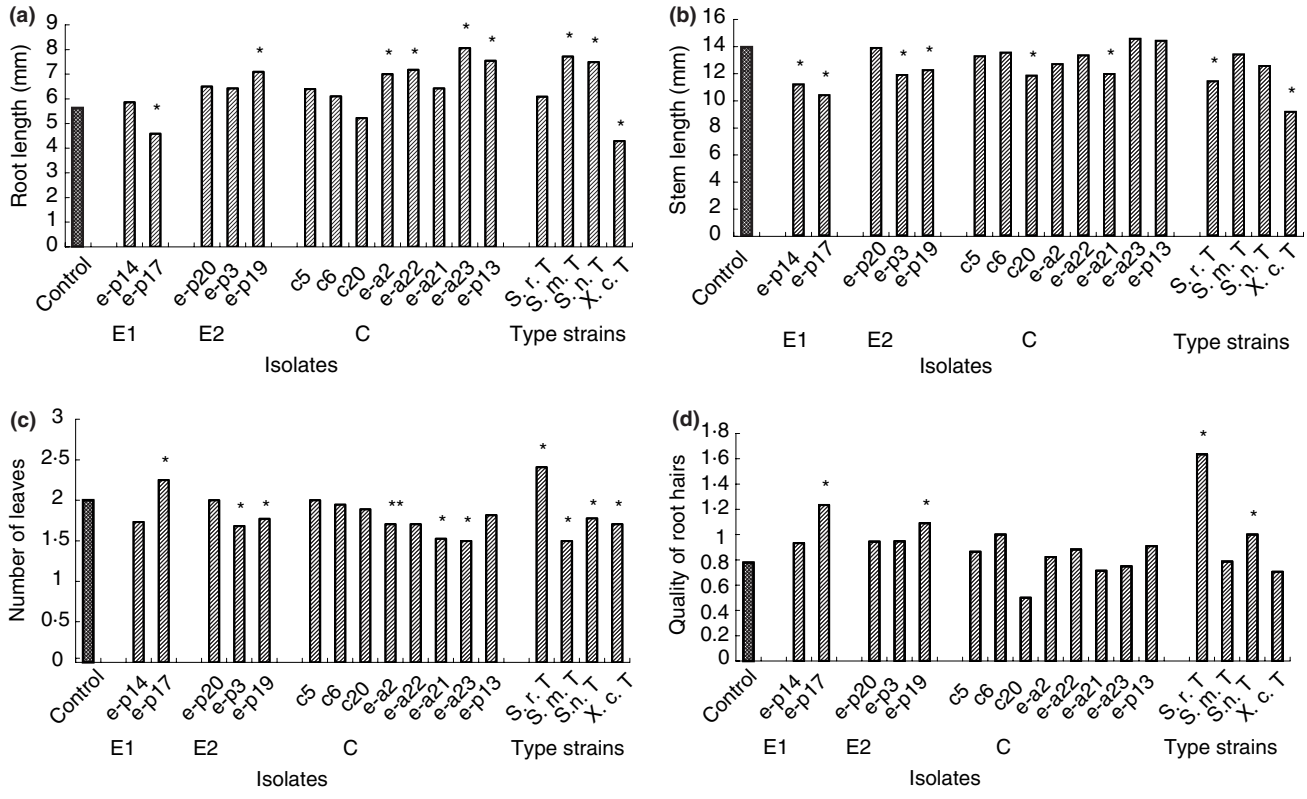

Fig. 1 Influence of the treatment of different *Stenotrophomonas* strains 5 weeks after treatment on (a) root length, (b) stem length, (c) the number of strawberry first leaves and (d) quality of roots compared with a nontreated control in a phytochamber assay. About 10 μl of a suspension of *Stenotrophomonas* at concentrations of 10^5 CFU ml^{-1} was applied on each of the 24 seeds per treatment using seed dipping for 15 min. Each experiment was repeated three times. Significant differences ($P \leq 0{\cdot}05$) were determined by Mann–Whitney and are indicated by asterisks. Type strains: S. r., *S. rhizophila* DSM 14405^T; S. m., *S. maltophilia* DSM 50170^T; S. n., *S. nitritireducens* DSM 12575^T and X. c., *Xanthomonas campestris* DSM 3586^T

on root hair development, increasing lengths by 110 and 59%. The phytopathogenic control *X. campestris* showed negative effects on both parameters. Therefore, the main positive influence of the *Stenotrophomonas* isolates was seen on the root, while more negative effects were observed for stem length and leave development. Further, when reviewing all parameters, the environmental strains DSM 14405, e-p17, e-p19 and the type strain *S. nitritireducens* exert the highest plant growth promotion effects, and the negative influence of *Xanthomonas* treatment on plant growth was confirmed.

After analysing averaged effect by the *Stenotrophomonas* genomovars of all parameters tested, environmental groups were seen to promote the highest plant growth (Table 2). Compared with the untreated control, isolates of genomovar E2 and C were able to enhance root length by 20%, whereas no cluster group affected total plant length. The isolates of genomovar E1 positively influenced leaf development and simultaneously, enhanced root hair development. Isolates of genomovar E2 were also able to enhance root hair development, while isolates of the C cluster statistically significantly reduced this parameter.

Dose-dependent effect of *Stenotrophomonas* treatment

In a second step, the dose-dependent effect of *Stenotrophomonas* treatment was analysed. Here, the type strains of *S. maltophilia* and *S. rhizophila* as well as strain *S. maltophilia* e-p20 were investigated in detail. For each strain, the effect of *Stenotrophomonas* treatment on plant growth was dependent on the bacterial population applied on germ buds (Fig. 2). Fig. 2a shows the influence of different bacterial concentrations on root length. For DSM 50170 and e-p20, an optimal concentration of 10^5 CFU ml^{-1} for root growth stimulation was found; at lower and higher concentrations, the growth decreased. Fig. 2b, which shows the effect on stem length, presents a similar picture. The two other parameters were also dose-dependent (Fig. 2c,d). At a concentration of 10^5 CFU ml^{-1}, *S. rhizophila* DSM

Table 2 Effect of *Stenotrophomonas* treatment on plant growth and production of indole-3-acetic acid analysed for genomovars and type strains

Cluster/strain	Influence on				Average IAA concentration (μg ml^{-1})
	Root length	Stem length	Leaves	Root hairs	
E1	n.s.	−	n.s.	+	3·4
E2	+	−	−	+	3·4
C	+	n.s.	−	+	1·5
DSM 50170^{T}	+	n.s.	−	−	1·1
DSM 14405^{T}	−	−	+	n.s.	4·1
DSM 12575^{T}	+	n.s.	−	+	3·1
DSM 3586^{T}	−	−	−	−	0

+, positive; −, negative, statistically significant; n.s., not statistically significant.

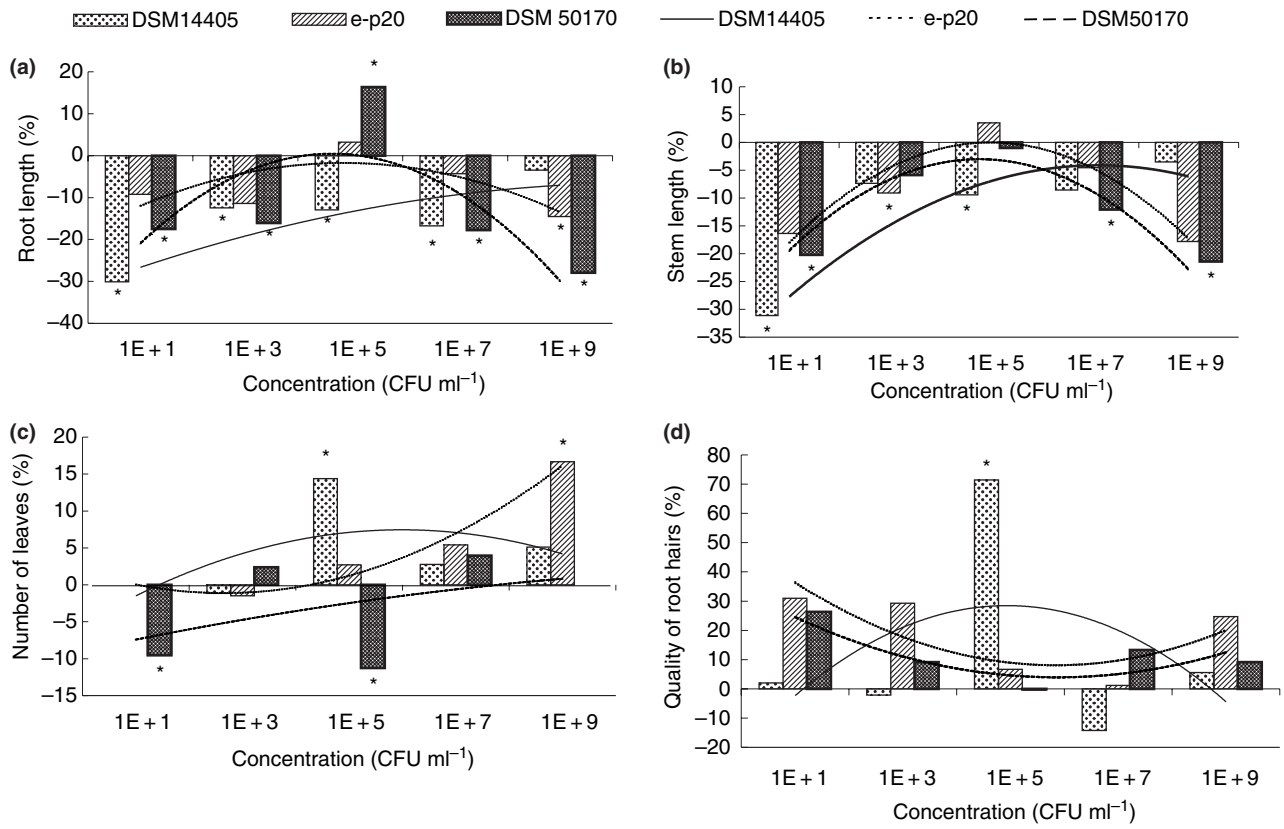

Fig. 2 Influence of different concentrations of three *Stenotrophomonas* strains (*S. maltophilia* e-p20, *S. rhizophila* DSM 14405, *S. maltophilia* DSM 50170) 5 weeks after treatment on (a) root length, (b) stem length, (c) the number of strawberry first leaves and (d) quality of roots compared with a nontreated control in a phytochamber assay. About 10 μl of a suspension of *Stenotrophomonas* at concentrations of 10^3, 10^5, 10^7 and 10^9 CFU ml^{-1} was applied on each of the 24 seeds per treatment using seed dipping for 15 min. Each experiment was repeated three times. Significant differences ($P \leq 0{\cdot}05$) were determined by Mann–Whitney and are indicated by asterisks

14405 was able to enhance root hair development by 71%. For the other two isolates, two optima were recognized. The dose effect for the increase of root hairs was statistically significant. Significant differences in leaf number were found for different *Stenotrophomonas* concentrations applied.

IAA production by *Stenotrophomonas* strains

The phytohormone IAA is often responsible for plant growth promotion induced by plant-associated bacteria. Therefore, the ability to produce IAA *in vitro* by *Stenotrophomonas* strains was investigated and all isolates were found to

Table 3 Bacterial production of indole-3-acetic acid (IAA) by *Stenotrophomonas* strains *in vitro*

Isolate	IAA concentration (μg ml^{-1})	IAA production*
e-p14	2·4	++
e-p17	3·7	+++
e-p20	3·2	+++
e-p13	1·9	++
e-p19	5·2	+++
c5	2·3	++
c6	0·9	+
c20	0·9	+
e-a2	1·3	+
e-a22	2·0	++
e-a21	2·2	++
e-a23	0·7	+
e-p13	2·5	++
DSM 50170^T	1·1	+
DSM 14405^T	4·1	+++
DSM 12575^T	3·1	+++
DSM 3586^T	0	−

*Assessment: +, 0.7–1.5 μg ml^{-1}; ++, 1.6–3 μg ml^{-1}; +++, 3 μg ml^{-1}; −, no IAA production.

produced it (Table 3). Concentrations ranged between 5·2 μg ml^{-1} for *S. maltophilia* e-p19 and 0·7 μg ml^{-1} for the aquatic strain e-a23. On average, the isolates of the environmental clusters E1 and E2 produced statistically significantly more IAA (3·4 μg ml^{-1}) than the isolates belonging to the clinical cluster (1·5 μg ml^{-1}, Table 2). In contrast to the phytopathogenic control, *X. campestris*, the type strain of *S. nitritireducens* also produced this phytohormone.

DISCUSSION

Three important objectives in *Stenotrophomonas* research will be discussed: (i) the importance of the dose-dependent effects for biocontrol applications, (ii) the impact of being able to differentiate between strains, and (iii) taxonomical questions. Members of the γ-proteobacterial genus *Stenotrophomonas* are of biotechnological interest as they produce antifungal antibiotics like maltophilin (Jacobi *et al.* 1996), alteramid (Berg *et al.* 1999) and xanthobaccin (Nakayama *et al.* 1999) along with a broad spectrum of extracellular enzymes like chitinases, proteases and glucanases (Minkwitz and Berg 2001). Many of these metabolites are responsible for biotechnological applications in bioremediation and in biological control of plant pathogens. The latter is an important alternative to chemical plant protection and a key element in environmentally friendly agriculture. Numerous studies have demonstrated the ability of *Stenotrophomonas* strains to suppress diseases caused by fungal plant pathogens (Berg *et al.* 1994; Kobayashi *et al.* 1995; Nakayama *et al.* 1999). One of the difficulties in developing rhizobacteria as a viable alternative is that many biological control agents are found to be too variable in their performance. According to Raaijmakers and Weller (2001) variable expression of genes involved in disease suppression and poor root colonization are the major factors contributing to this inconsistency. Here, we showed a clear dose-dependent effect of the *Stenotrophomonas* treatment; this is important for biocontrol applications. Only a specific concentration for each strain could enhance plant growth, while lower and higher concentrations resulted in plant growth reduction. One reason for this dose-dependent effect could be the regulation of secondary metabolites. This phenomenon subject to complex regulation, allowing bacteria to sense their own population density by the production of *N*-acyl-L-homoserine lactones (AHLs) and to respond to different environmental factors (Eberl 1999). However, nothing is known about 'quorum sensing' mechanisms in *S. maltophilia* and *S. rhizophila*. Production of AHLs by *Stenotrophomonas* isolates, which was investigated by the aid of the bioluminescent sensor plasmid pSB403 (Winson *et al.* 1998) showed no production of these substances (L. Eberl and G. Berg, unpublished data).

However, bacterium–plant interactions can be mediated by the synthesis of plant hormones, e.g. IAA. For all *Stenotrophomonas* strains, IAA production *in vitro* was shown although in different amounts. Interestingly, isolates of genomovar C, which contains clinical and aquatic strains, produce significantly lower amounts than plant-associated strains. This group of strains also showed a significantly lower effect in plant growth promotion tested in our microtitre plate assay than plant-associated strains. The strain e-p19 which produced the highest amounts of IAA *in vitro* showed negative effects on stem length and positive effects on root length and root hair development. Similar effects were found for other isolates with high *in vitro* production of IAA. The plant hormone IAA (auxin) is widely distributed in the plant kingdom and has been linked to root formation and stem growth promotion; however, the actual pathway of its action is still only partially known. For *Xanthomonas*, no IAA production could be found. Hence, IAA cannot be responsible for the concentration-dependent effect. This was confirmed by genome analysis of *X. campestris* as no homologues to genes encoding proteins for IAA synthesis were identified (Van Sluys *et al.* 2002). The rhizosphere, the microenvironment of plant where *Stenotrophomonas* mostly occurs (Juhnke and Des Jardins 1989; Berg *et al.* 1996), is a microbial hotspot where a high diversity of associated micro-organisms can be found. Our results were obtained by *in vitro* investigations, but one should bear in mind that *ad planta* the situation is much more complex due to the influence of biotic and abiotic

factors. However, in other studies using the strawberry seedling assays to observe plant growth effects by rhizobacteria, a correlation between effects observed *in vitro* and *ad planta* was found (Berg *et al.* 2001).

A second point of interest is to distinguish between clinical and environmental *Stenotrophomonas* isolates (Berg *et al.* 1999). Using various molecular fingerprinting methods, high heterogeneity was observed in *Stenotrophomonas*, although it was not possible to differentiate between clinical and environmental isolates. Minkwitz and Berg (2001) differentiated the genomovars by 16S rDNA sequence analysis. Using this method, it was possible to divide plant-associated from clinical strains, but not aquatic from clinical strains. Our results show that clinical isolates of *Stenotrophomonas* are also able to influence plant growth, although to a lesser extent than plant-associated strains. It could be that these strains' original microenvironment was plant and that later they spread into hospitals, where they lost some of their abilities not necessary for survival under the new conditions.

Stenotrophomonas and *Xanthomonas* are phylogenetically closely related (Nesme *et al.* 1995; Hauben *et al.* 1997). However, *S. maltophilia* was formerly classified as *X. maltophilia*, but because of significant physiological and genotypic differences to other *Xanthomonas* strains, a new genus was created (Palleroni and Bradbury 1993). Moore *et al.* (1997) found a 3% (45–68 nt) difference in 16S rDNA sequences between the two genera. In an earlier study, we obtained 48 different nucleotides of 1534 between the *S. maltophilia* type strain DSM 50170 and *X. campestris* XCC15, which also corresponds exactly to 3%. Normally, sequence differences of about 3% suggest that strains belong to different species, not genera. Both *Stenotrophomonas* and *Xanthomonas* are known for their interaction with eucaryotic organisms. *Stenotrophomonas* strains cause fatal infections in immunocompromised humans, but are positive with plants due to their ability to enhance plant growth and suppress plant pathogens. However, *Xanthomonas* species are important plant pathogens of a broad host range. Here, we show, for the first time, that at higher concentrations, *Stenotrophomonas* strains are also able to damage plants. Therefore, the *Stenotrophomonas*–plant interaction was highly sensitive to *Stenotrophomonas* concentration which determined whether positive or negative effects were found.

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