Barakacin: A Thiazolyl-indole Alkaloid Isolated from Ruminal *Pseudomonas* sp.

Imene Zendah [a], Khaled A. Shaaban [a], Elisabeth Helmke [b], Armin Maier [c], Heinz H. Fiebig [c], and Hartmut Laatsch [a,*]

[a] *Institute of Organic and Biomolecular Chemistry, Georg-August University, Tammannstrasse 2, D-37077 Göttingen, Germany*
[b] *Alfred-Wegener-Institute for Polar and Marine Research, Am Handelshafen 12, D-27570 Bremerhaven, Germany*
[c] *Oncotest GmbH, Am Flughafen 12-14, D-79108 Freiburg, Germany*

Figure S1. Working up scheme for *Pseudomonas aeruginosa*.

Figure S2. TLC of barakacin (**1**) on silica gel, developed with CH_2Cl_2/ 2% MeOH. **A**: barakacin (**1**) under UV light at 366 nm; **B**: after spraying with anisaldehyde/H_2SO_4; **C**: after spraying with Ehrlich's reagent.

Figure S3. ^{1}H NMR spectrum (300 MHz, $CDCl_3$) of Barakacin (**1**).

Figure S4. ^{13}C NMR spectrum (125 MHz, $CDCl_3$) of Barakacin (**1**).

Figure S5. ^{1}H-^{1}H COSY spectrum (600 MHz, $CDCl_3$) of Barakacin (**1**).

Figure S6. HMBC spectrum (600 MHz, $CDCl_3$) of Barakacin (**1**).

Figure S7. ^{1}H-^{1}H COSY (bold bonds) and (→) HMBC correlations in partial structures A–C of Barakacin (**1**).

Figure S8. Antimicrobial activity of supernatant from liquid culture of ZIO against *Streptococcus* sp. R36 (**A**) and *Staphylococcus aureus* R25 (**B**), tested by agar well diffusion method.

Table S1. Antimicrobial activity of the ruminal bacterium *Pseudomonas aeruginosa* strain ZIO against a collection of human-pathogenic bacteria.

∗ Corresponding author. Tel: +49-551-393211. Fax: +49-551-399660.
E-mail: hlaatsc@gwdg.de

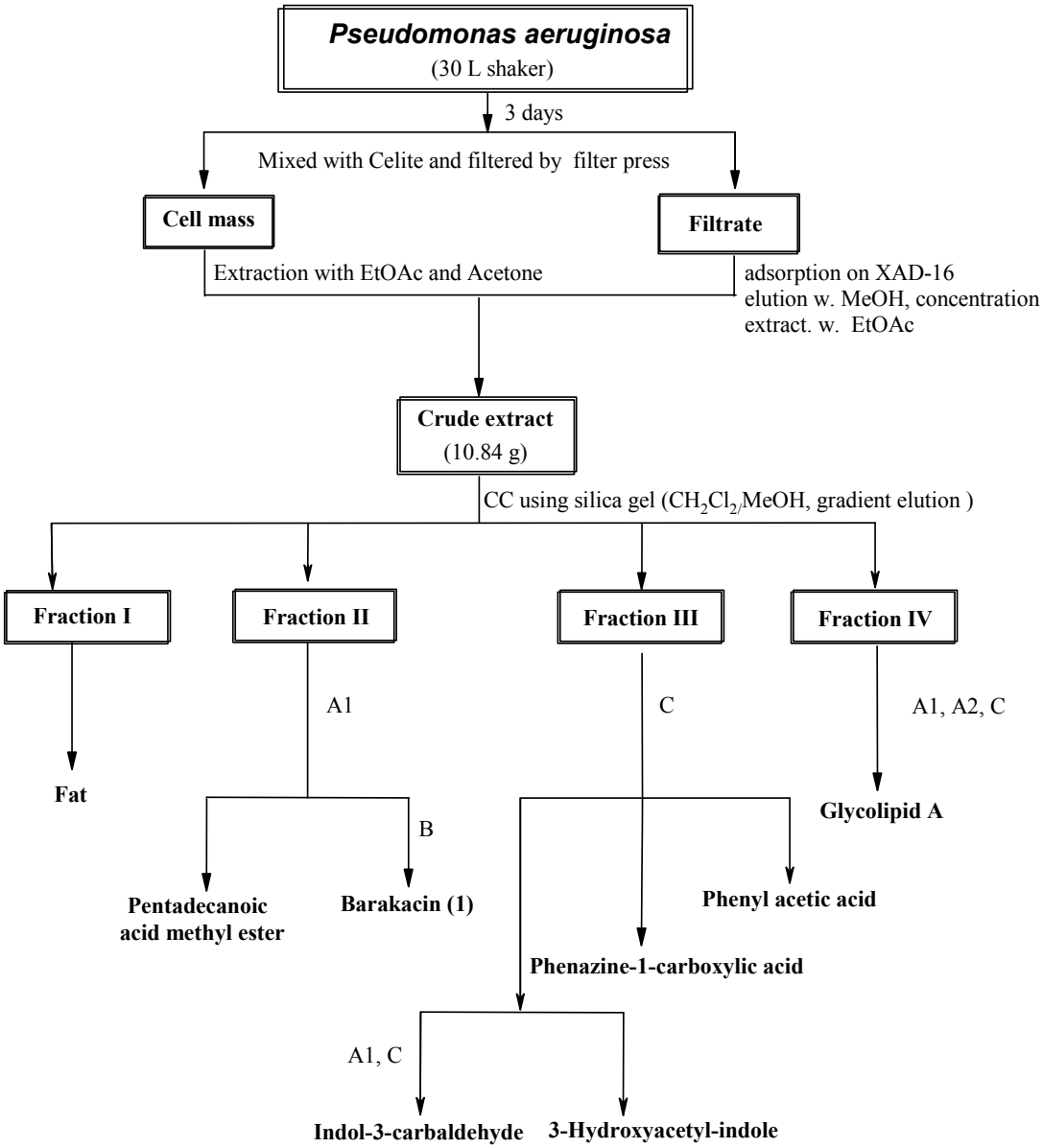

Figure S1. Working up scheme for the ruminal bacterium *Pseudomonas aeruginosa*.

A1 = Sephadex LH-20 (CH_2Cl_2/ 40% MeOH)

A2 = Sephadex LH-20 (MeOH)

B = PTLC (CH_2Cl_2/ 5% MeOH)

C = Silica gel column (CH_2Cl_2/MeOH)

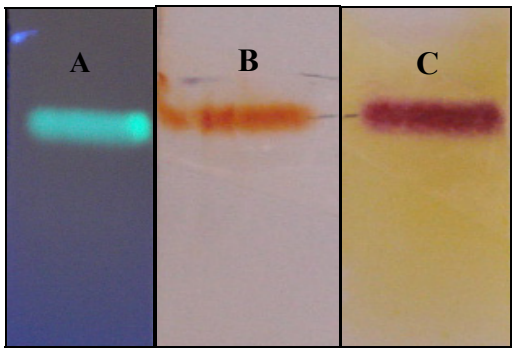

Figure S2. TLC of barakacin (**1**) on silica gel, developed with CH_2Cl_2/ 2% MeOH. **A**: barakacin (**1**) under UV light at 366 nm; **B**: after spraying with anisaldehyde/H_2SO_4; **C**: after spraying with Ehrlich's reagent.

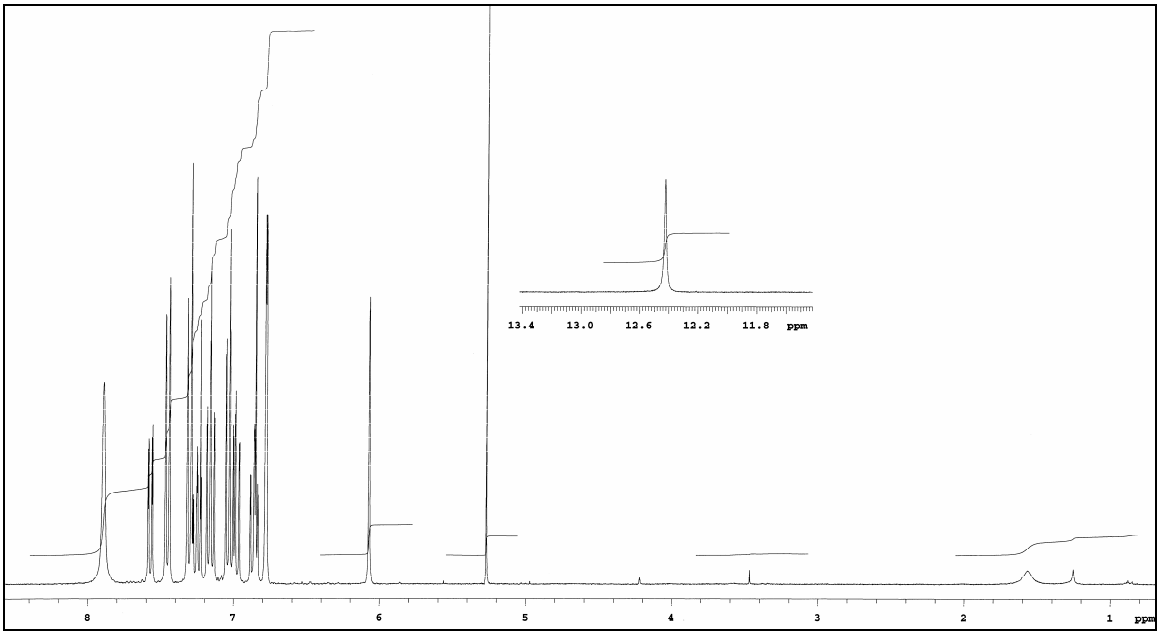

Figure S3. ^{1}H NMR spectrum (300 MHz, $CDCl_3$) of Barakacin (**1**).

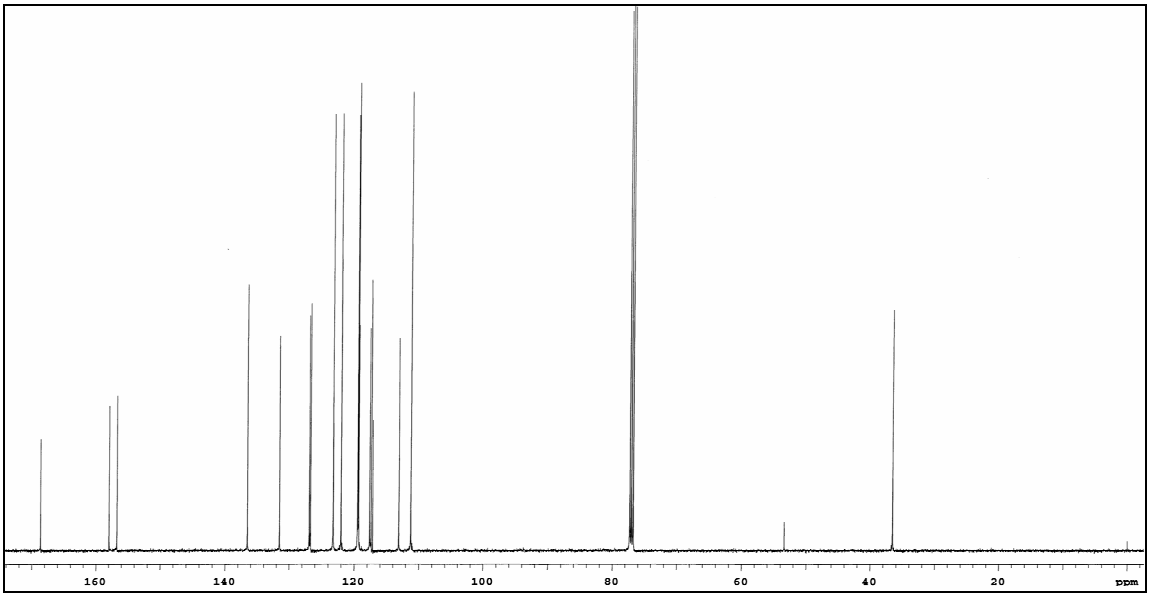

Figure S4. ^{13}C NMR spectrum (125 MHz, $CDCl_3$) of Barakacin (**1**).

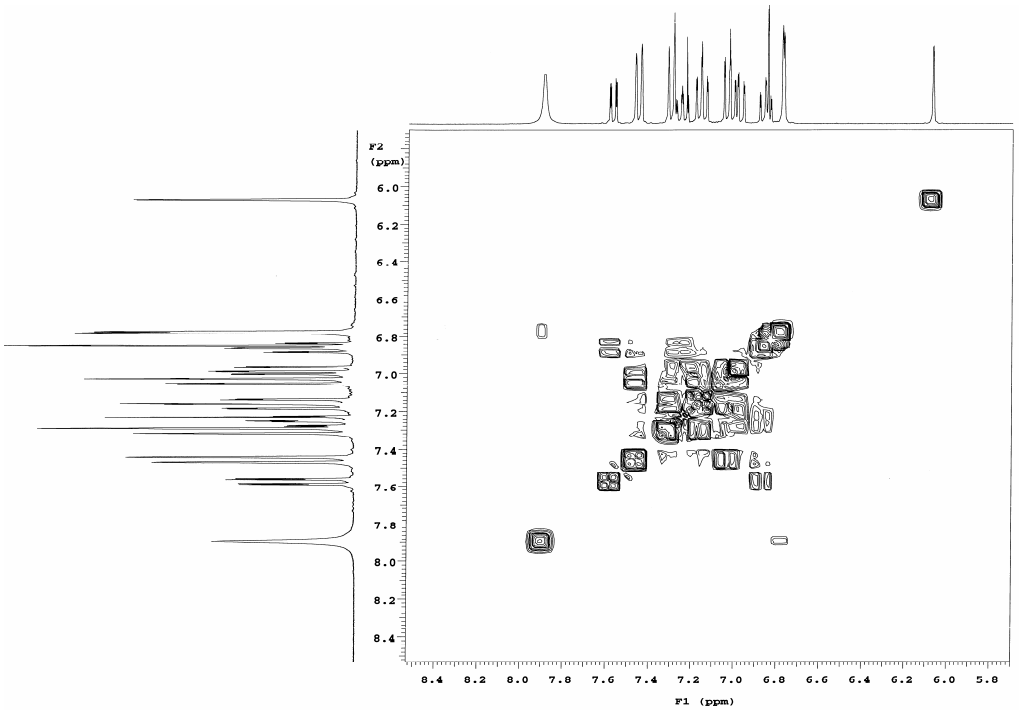

Figure S5. ^{1}H-^{1}H COSY spectrum (600 MHz, $CDCl_3$) of Barakacin (**1**).

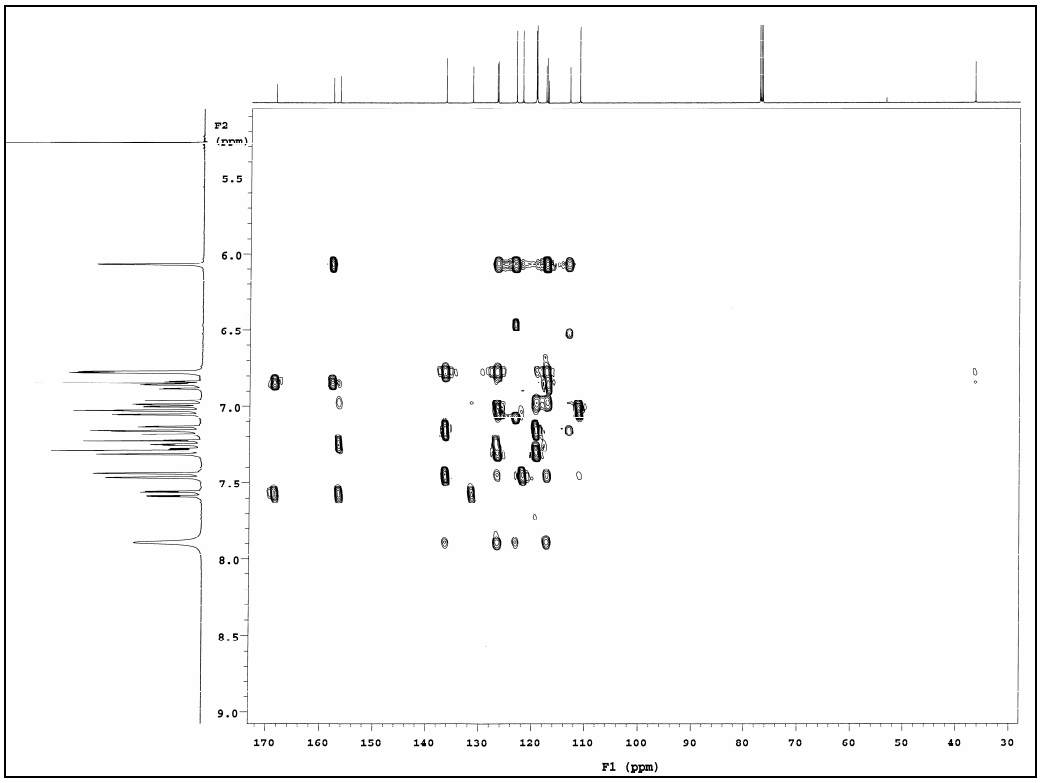

Figure S6. HMBC spectrum (600 MHz, $CDCl_3$) of Barakacin (**1**).

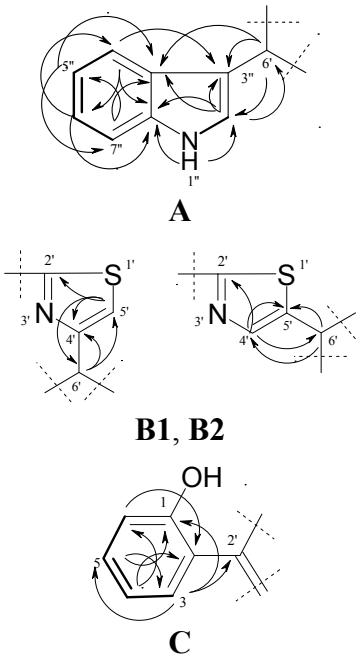

Figure S7. ^{1}H-^{1}H COSY (bold bonds) and (→) HMBC correlations in partial structures A–C of barakacin (**1**).

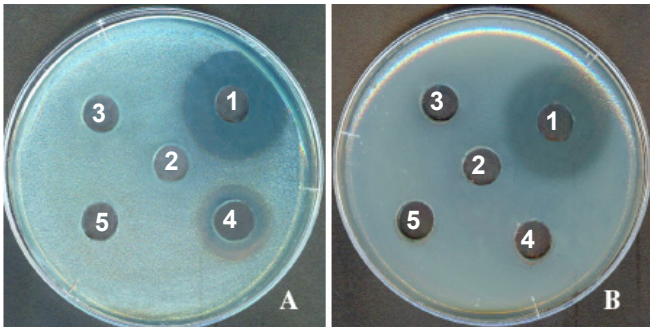

Figure S8. Antimicrobial activity of supernatant from liquid culture of ZIO against *Streptococcus* sp. R36 (**A**) and *Staphylococcus aureus* R25 (**B**), tested by agar well diffusion method. For comparison, a ruminal *Proteus vulgaris* strain was tested.

1: supernatant from liquid culture of ZIO
2: liquid PY medium (positive control)
3: cell mass from liquid culture of ZIO
4: supernatant from liquid culture of the ruminal bacterium *Proteus vulgaris* (Sa14) (moderate activity, 12 mm, against *Streptococcus* sp. (R36) (**A**) and inactive against *Staphylococcus aureus* (R25) (**B**))
5: cell mass from liquid culture of the ruminal bacterium *Proteus vulgaris* (Sa14)

Table S1. Antimicrobial activity of the ruminal *Pseudomonas aeruginosa* ZIO against a collection of human-pathogenic bacteria. Tested was the supernatant of the culture grown on PY medium.

Target References[a)]	Genera and species	Samples	Department	Activity of strain ZIO [b)]
R1	*Enterobacter aerogenes*	urine	emergency	++
R2	*Enterobacter aerogenes*	pus	cardiovasculary	-
R3	*Enterobacter aerogenes* BLSE	urine	neonatal	++
R4	*Serratia* BLSE	pus	reanimation	++
R5	*Serratia liquefaciens*	urine	urology	+++
R6	*Staphylococcus aureus*	hemoculture	reanimation	+++
R7	*Providencia* BLSE	visceral sound	visceral chirurgy	++
R8	*Staphylococcus aureus* MRSA	pus	maternity	+++
R9	*Staphylococcus* coagulase-negative	hemoculture	urology	+++
R10	*Staphylococcus aureus* MRSA	pus	dermatology	+++
R11	*Staphylococcus aureus* MRSA	lung	reanimation	+++
R12	*Staphylococcus aureus*	pus	urology	+++
R13	*Staphylococcus aureus*	pus	dermatology	+++
R14	*Staphylococcus aureus*	pus	cardiology	+++
R15	*Staphylococcus aureus*	nose swab	neonatal	+++
R16	*Staphylococcus aureus*	lung	reanimation	-
R17	*Staphylococcus* coagulase-negative	hemoculture	cardiology	+++
R18	*Xanthomonas maltophilia*	lung	neonatal	-
R19	*Staphylococcus* coagulase-negative	hemoculture	reanimation	+++
R20	*Xanthomonas maltophilia*	catheter	neonatal	-
R21	*Xanthomonas maltophilia*	-	reanimation	+++
R22	*Proteus mirabilis* BLSE	catheter	reanimation	+++
R23	*Staphylococcus aureus*	hemoculture	neonatal	+++
R24	*Ochrobacterium anthropi*	anal	neonatal	+++
R25	*Staphylococcus aureus*	hemoculture	neonatal	+++
R26	*Escherichia coli*	pus	urology	+++
R27	*Escherichia coli*	pus	visceral chirurgy	+++
R28	*Escherichia coli*	lung	reanimation	++
R29	*Escherichia coli* (BLSE)	urine	emergency	+
R30	*Staphylococcus* coagulase-negative SCN	hemoculture	reanimation	+++
R31	*Staphylococcus* coagulase-negative SCN	hemoculture	urology	+++
R32	*Escherichia coli*	anal	reanimation	+++
R33	*Escherichia coli*	urine	maternity	+++
R34	*Escherichia coli*	rectal	reanimation	+++
R35	*Klebsiella pneumoniae BLSE*	gastric swab	neonatal	+++
R36	*Streptococcus A*	pus	dermatology	+++

[a)] The R numbers refer to the lab codes of the test strains
[b)] Inhibition zone between 0 - 2 mm ≡ -; 2 - 12 mm ≡ +; 12 -20 mm ≡ ++; 20-50 mm ≡ +++.