SUPPLEMENTARY INFORMATION

Table S1. Related to Table 1. Inhibition of *C. scindens* and *Staph. aureus* by clinical strains of *C. difficile*

Clinical ***C. difficile*** (ATCC Number)	**Inhibitory test**		Toxinotype	Binary Toxin	Ribotype	Isolation source
	C. scindens ATCC 35704 **(0.75 O.D/PYF)** †	*Staph. aureus* MRSA **(1.2 O.D/PYF)**				
ATCC 9689	0.08±0.04‡*	0.23±0.15*	0	ND	001	clinical isolate (Type strain)
BAA-1870	0.04±0.01*	0.06±0.02*	IIIb	Yes	027	clinical isolate (ME, US)
BAA-1801	0.10±0.02*	0.54±0.11*	tcdA-, tcdB-	ND	010	human Feces (Belgium)
BAA-1814	0.04±0.01*	0.04±0.01*	XXII	Yes	251	clinical Isolate (unknown)

†:O.D at fresh PYF broth as control, ‡: Average±SD. *: Significant growth inhibition, $p<0.05$

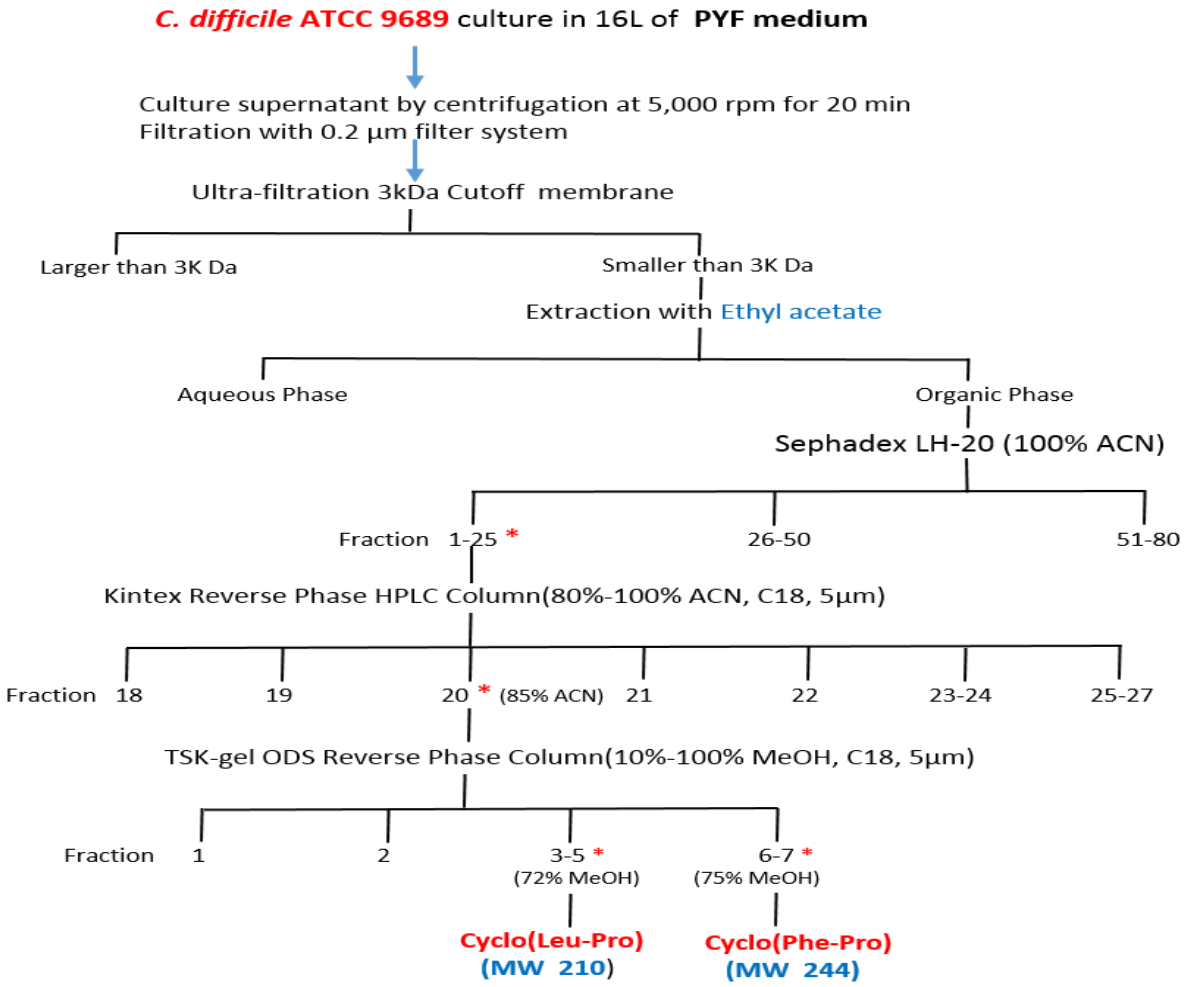

Figure S1. Related to Figure 2. Purification protocol for isolating antibacterial compounds secreted by *Clostridium difficile* ATCC 9689. The purification protocol started with 16 liters of spent PYF culture medium (24 hours incubation) of *C. difficile* ATCC 9689. The medium was initially filter sterilized using a 0.2 micron filter, followed by ultrafiltration through a 3 kDa filter. The filter sterilized and size fractionated supernatant fluid was extracted with ethyl acetate, followed by Sephadex LH 20 column chromatography using 100% ACN solvent, followed by two sequential C-18 reverse-phase HPLC solvent systems. Each fraction was analyzed for antibacterial activity by measuring inhibition of the growth of *Clostridium scindens* ATCC 35704 in PYF medium. Purified fractions (3 to 5 and 6 and 7) from the final HPLC step, with antibacterial activity, were then submitted for LC-ESI/MS and ^{1}H and ^{13}C NMR analysis (STAR Methods).

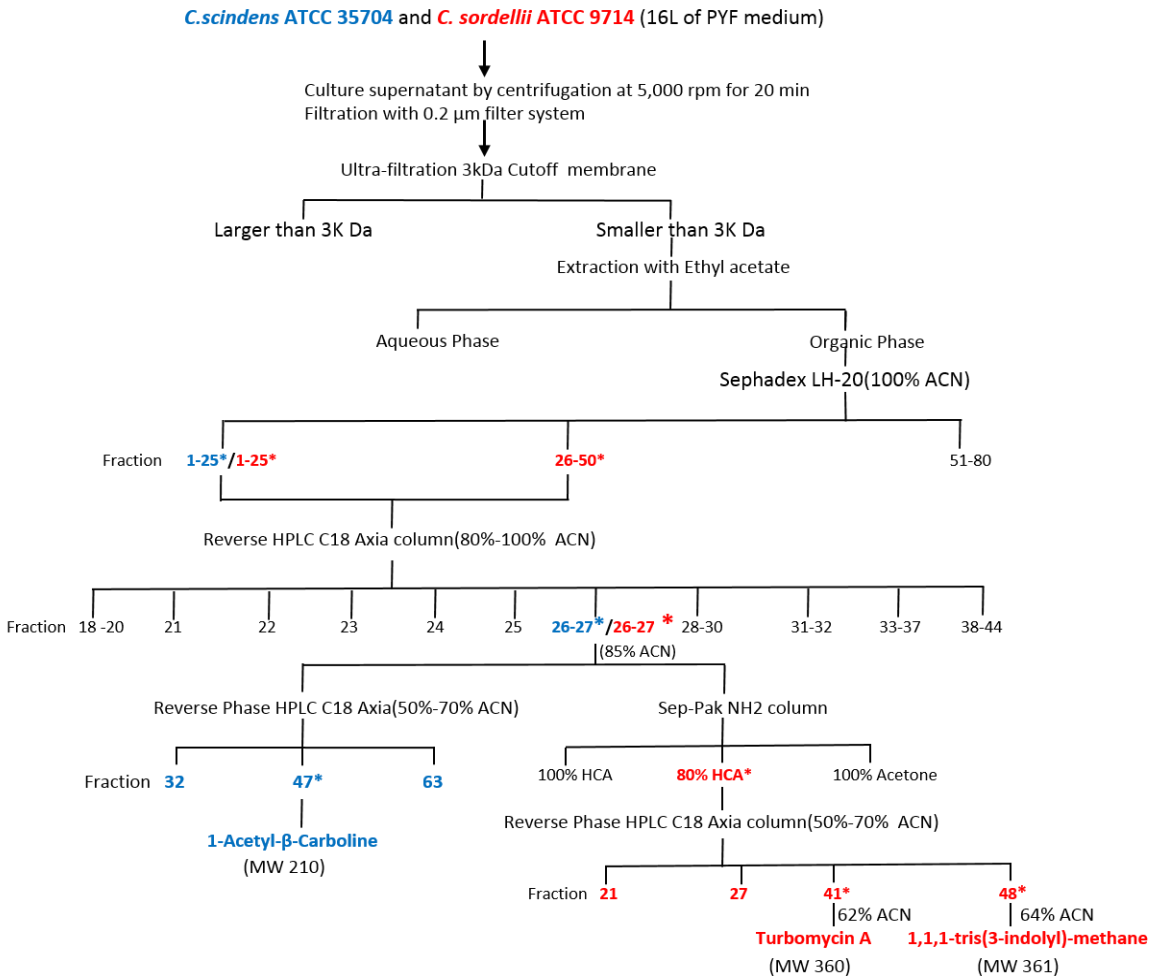

Figure S2. Related to Figure 3. Purification of antibacterial compounds secreted by *Clostridium scindens* (blue) and *Clostridium sordellii* (red). Purification of compounds from each bacterium started with 16 liters of spent PYF culture medium (24 hrs.). The purification began with filter sterilization through a 0.2µ filter followed by a 3 kDa filter. The compound(s) were then extracted into ethyl acetate followed by Sephadex LH-20 column chromatography using 100% ACN solvent. A series of C-18 reverse phase HPLC solvent systems were used to purify each compound until a single symmetrical peak was obtained. Each fraction was assayed for antibacterial activity using *Clostridium difficile* ATCC 9689 as test bacterium. The pure compounds were then sent for LC-ESI/MS and ^{1}H and ^{13}C NMR analysis (STAR Methods).

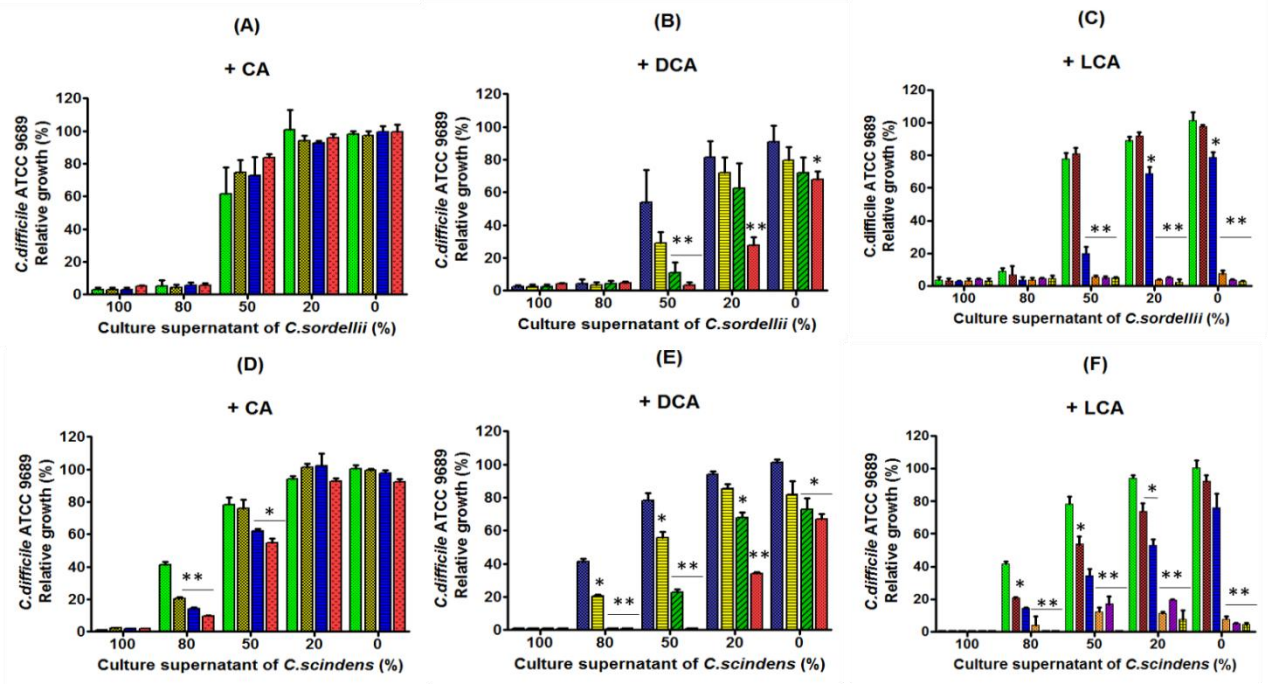

Figure S3. Related to Figures 4 and 5. Effect of bile acid addition to spent culture medium of *C. sordellii* and *C. scindens* on growth of *C. difficile*. Spent culture PYF medium (24 hrs.) from either *C. sordellii* or C. *scindens* was filter sterilized, pH adjusted to 7,2, diluted with increasing amounts of fresh PYF medium until growth was approximately 40% to 60% maximal. Varying concentrations of either cholic acid (CA), No CA, 25uM CA, 50uM CA, 100uM CA or deoxycholic acid (DCA), No DCA, 25uM DCA, 50uM DCA, 100uM DCA or lithocholic acid, No LCA, 1.25uM LCA, 5.0uM LCA, 12.5uM LCA, 25uM LCA, 50uM LCA was added to diluted spent culture media,

inoculated with 10^6/ml vegetative cells of *C. difficile*, incubated for 24 hrs, and OD readings measured at 660 nm. *p<0.05, ** p<0.01.

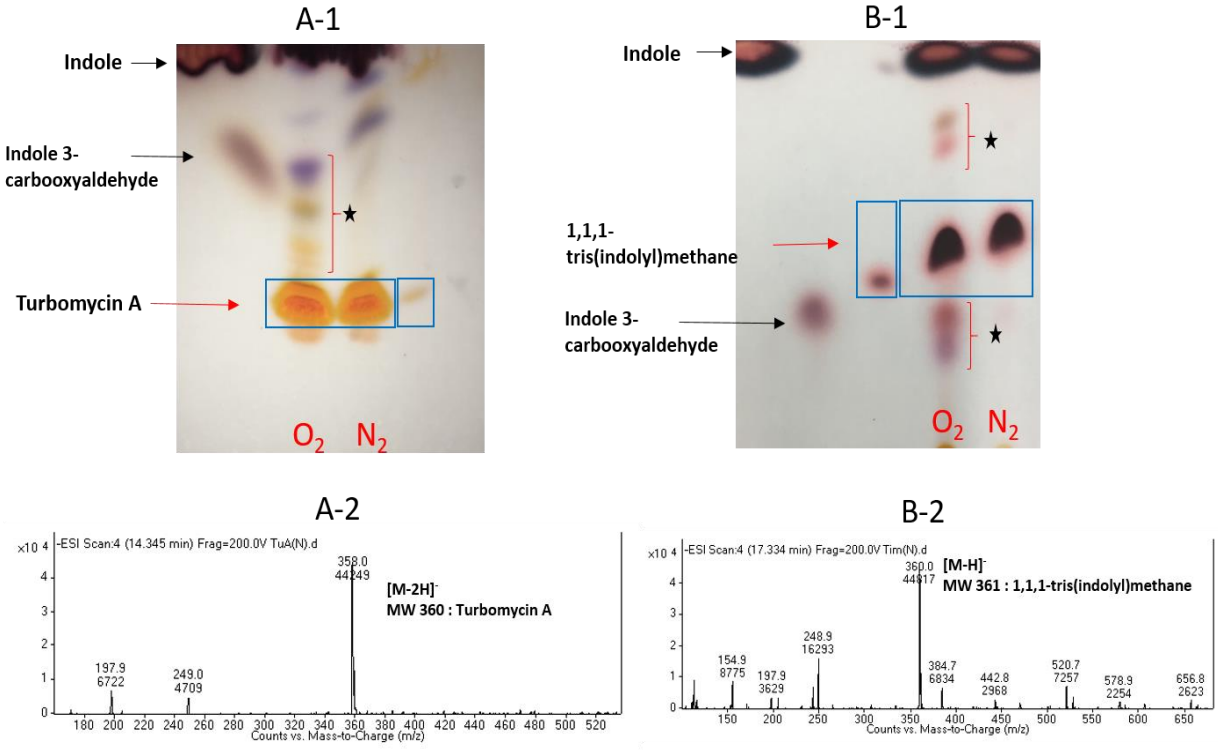

Figure S4. Related to Figures 4 and 5. Chemical synthesis turbomycin A and of 1,1,1,-tris(3-indolyl)-methane (TIM) under aerobic (O_2) vs anaerobic (N_2) conditions. (See STAR Methods). The reactants (indole and indole 3-carboxyaldehyde) and product (turbomycin A) was separated on TLC using a solvent system of (chloroform:methanol:toluene, 80:20:2, vol/vol/vol) left panel (**A-1**). The reactant substrates and product (1,1,1-tris-(3-indolyl)-methane, TIM) was separated on TLC using a solvent system of (chloroform:hexane:acetone, 60:20:10, vol/vol/vol) and products shown on the right panel (**B-1**). The substrates and products for each reaction were detected using a mixture of van Urk and Salkowski (1:3, vol/vol) reagent (Ehmann, 1977). *Unknown oxidized/degradation products. Product identification of purified chemically synthesized turbomycin A and TIM were determined by mass spectrometry (**A-2**) and TIM (**B-2**).

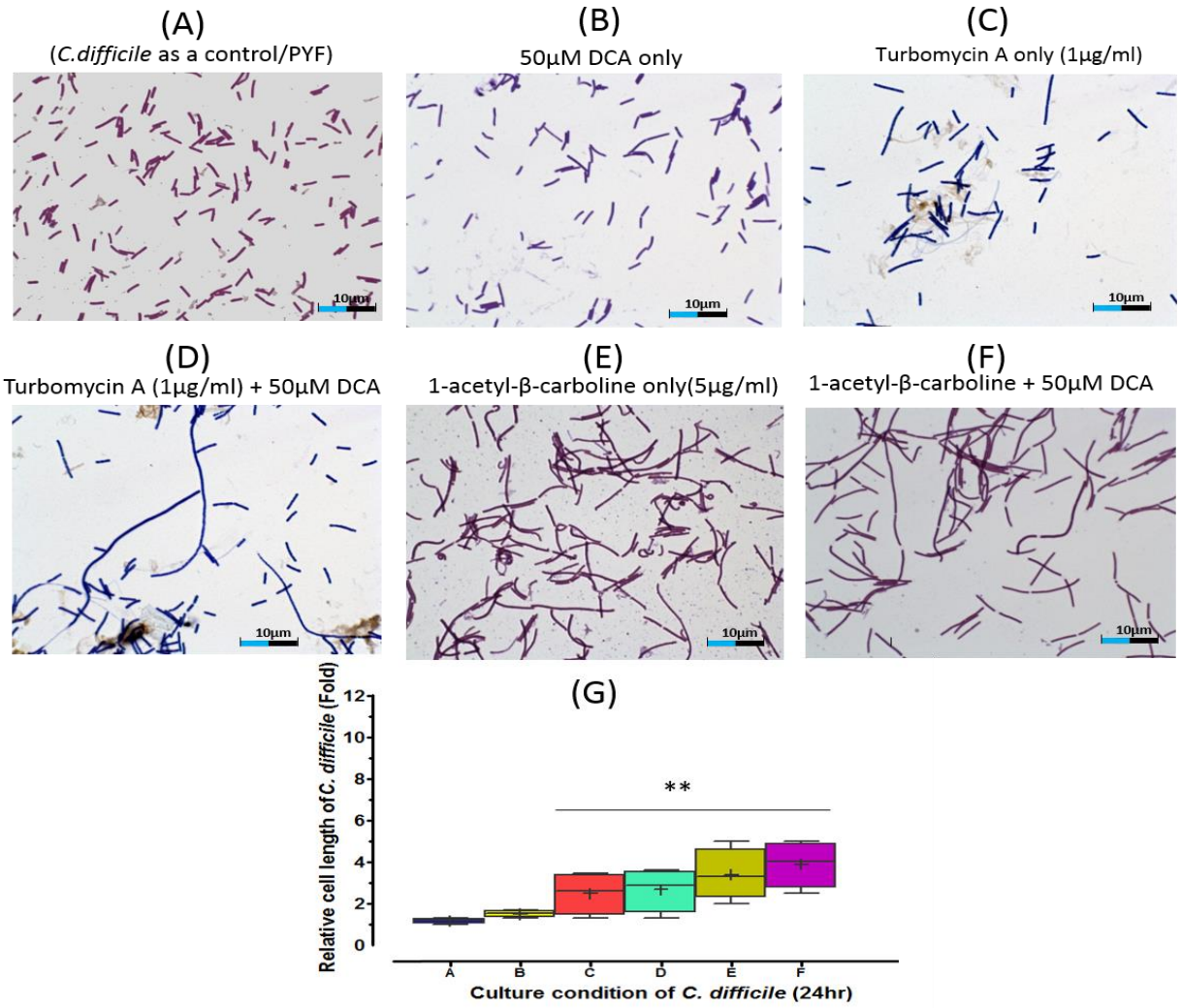

Figure S5. Related to Figure 6. Changes in *C. difficile* ATCC 9689 cell morphology induced by turbomycin A and 1-acetyl-β-carboline with and without deoxycholic acid (A-F). Vegetative cells 10^6 were inoculated into PYF medium containing: A) no additions; B) DCA (50 µM); C) turbomycin A (1 µg/ml); D) turbmycin A (1 µg/ml) + DCA (50 µM); E) 1-acetyl-β-carboline (5µg/ml) and F) 1-acetyl-β-carboline (5µg/ml) + DCA (50 µM) and incubated for 24 hrs. Each culture (1ml) was concentrated to 100 µl by centrifugation, 5 µl of cell suspension loaded on to a slide for Gram staining and photo images taken (Methods). Cell length was determined by using indicated size bar in several microscopic fields. (Magnification 1000x). ** P<0.01