

Supporting Information

TABLE S1. UPLC-HRMS analysis of the intermediates involved in anaerobic testosterone catabolism by *S. denitrificans*.

No. ^a	Compound ID	UPLC behavior (RT ^b , min)	Molecular formula/ (predicted molecular mass) ^c	Dominant ion peaks	Identification of product ions	Mode observed
1	Testosterone	6.20	C ₁₉ H ₂₈ O ₂ 288.2082	271.2062 289.2147 311.1987	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
1'	Androst-4-en-3,17-dione	5.96	C ₁₉ H ₂₆ O ₂ 286.1926	269.1909 287.2031 309.1831	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
2	1-Dehydrotestosterone	5.82	C ₁₉ H ₂₆ O ₂ 286.1926	269.1903 287.2000 309.1823	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
2'	Androsta-1,4-diene-3,17-dione	5.50	C ₁₉ H ₂₄ O ₂ 284.1770	267.1742 285.1852 307.1666	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
3	1-Testosterone	6.55	C ₁₉ H ₂₈ O ₂ 288.2082	271.2062 289.2163 311.1987	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
3'	Androst-1-en-3,17-dione	6.35	C ₁₉ H ₂₆ O ₂ 286.1926	269.1905 287.1999 309.1831	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
4	1,17-dihydroxy-androstan-3-one	5.64	C ₁₉ H ₃₀ O ₃ 306.2187	289.2168 307.2267 329.2093	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
5	17-hydroxy-androstan-1,3-dione	5.05	C ₁₉ H ₂₈ O ₃ 304.2013	287.2011 305.2127 327.1936	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
5'	Androstan-1,3,17-trione	4.72	C ₁₉ H ₂₆ O ₃ 302.1875	285.1855 303.1915 325.1780	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
6	17-hydroxy-1-oxo-2,3- <i>seco</i> -androstan-3-oic acid (2,3-SAOA)	5.42	C ₁₉ H ₃₀ O ₄ 322.2136	305.2117 323.2227 345.2042	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
6'	1,17-dioxo-2,3- <i>seco</i> -androstan-3-oic acid	5.34	C ₁₉ H ₂₈ O ₄ 320.1980	303.1960 321.2070 343.1885	[M-H ₂ O+H] ⁺ [M+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI
X	Compound X	4.22	C ₁₇ H ₂₆ O ₄ 294.1824	259.1705 277.1823 317.1718	[M-2H ₂ O+H] ⁺ [M-H ₂ O+H] ⁺ [M+Na] ⁺	ESI and APCI ESI and APCI ESI

^aCompound numbers mentioned in **Fig. 3A**. ^bRT, retention time. ^cThe predicated molecular mass was calculated using the atom mass of ¹²C (12.0000), ¹⁶O (15.9949), and ¹H (1.0078). The common initial intermediates involved both in aerobic and anaerobic catabolism are highlighted in bold.

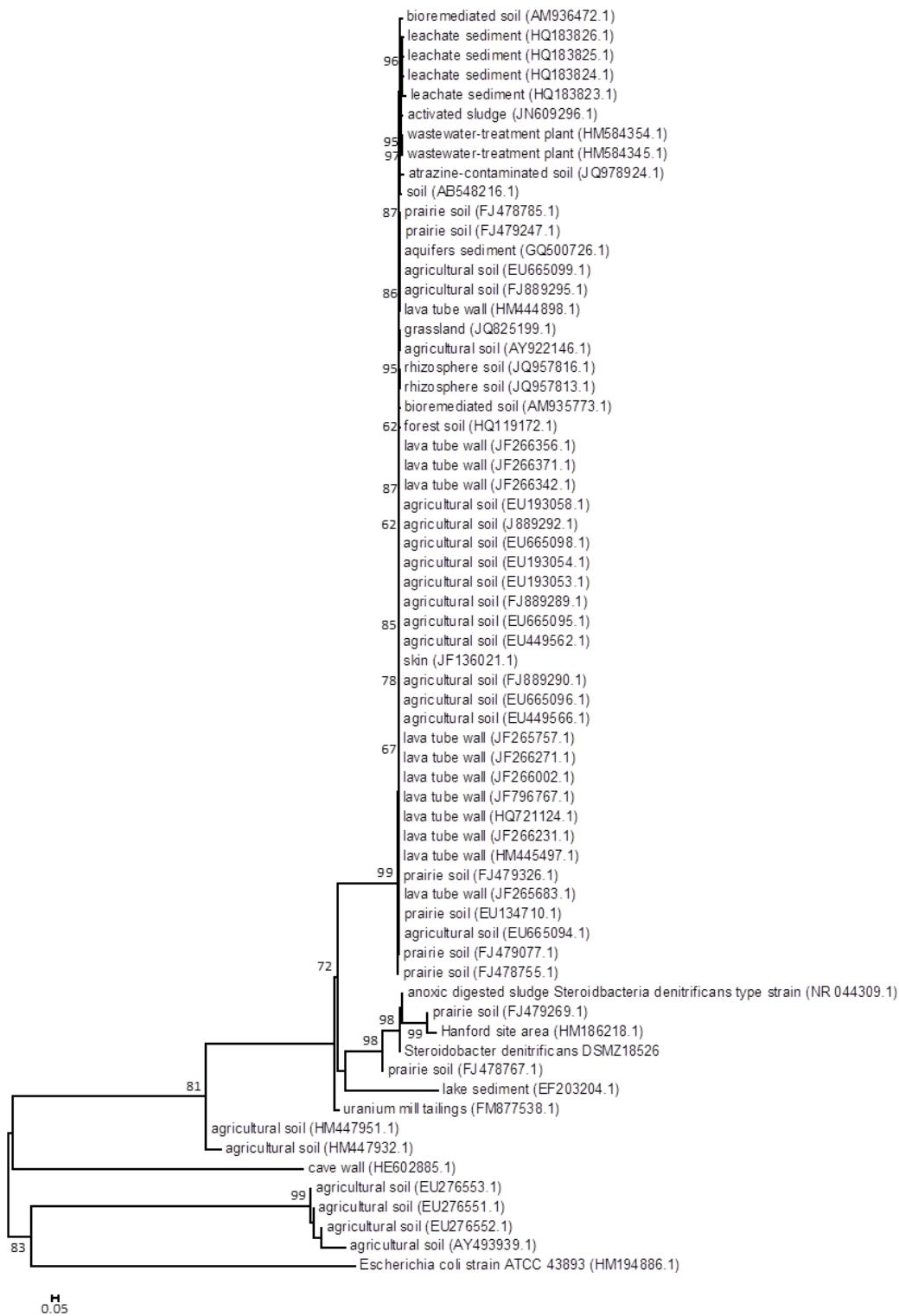


Fig. S1. Phylogenetic analysis of *S. denitrificans* strains. Sixty-three 16S rRNA genes were retrieved from NCBI with more than 98% nucleotide sequence similarity to the 16S rRNA gene of *S. denitrificans* DSMZ18526. 16S rRNA gene of *Escherichia coli* ATCC 43893 served as an outgroup sequence. The neighbor-joining consensus gene tree contains 65 genes. The bootstrap confident values were obtained with 1000 re-samplings. The scale bar indicates 0.05 substitutions per nucleotide position.