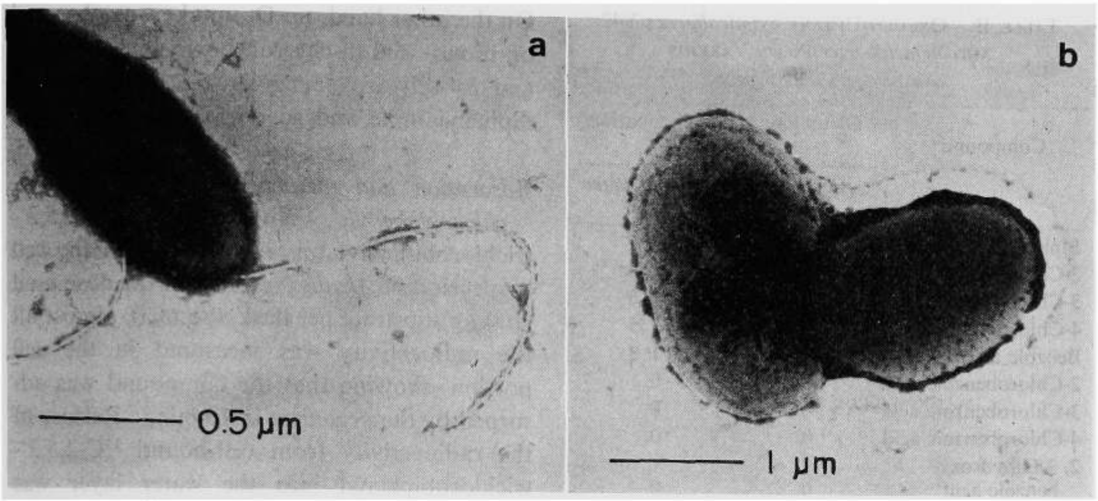

FIG. 1. Electronmicrographs of *Alcaligenes* Y42 (a) and *Acinetobacter* P6 (b).

ammonium salts, poly β-hydroxybutyrate: not produced. This organism was identified as a strain of *Acinetobacter*.

Growth and O_2 uptake on a variety of aromatic compounds

A variety of aromatic compounds were examined for growth and O_2 uptake of these two organisms. As shown in Table I, the both organisms grew well on biphenyl, 4-chlorobiphenyl and 4-methylbiphenyl. When *Acinetobacter* P6 was cultured with 4-chlorobiphenyl, a large amount of 4-chlorobenzoic acid was accumulated in the culture broth and the pH was dropped to 6.0 (initial pH 7.5) after 5 days incubation. Benzoic acid and protocatechuate also supported the growth. However, no growth was observed on mono- and dichlorobenzoic acids and 2,3- and 2,4-dihydroxybenzoic acid. 2-Phenylphenol, 4,4′-biphenol, diphenylamine, diphenylmethane, benzhydrol, benzophenone did not support the growth in both *Alcaligenes* Y42 and *Acinetobacter* P6 in spite of their similar structures to biphenyl.

As in the case of growth test, biphenyl was the best substrate for O_2 uptake of both organisms (Table II). Good O_2 uptake was observed in 2-, 3-, and 4-chlorobiphenyl, benzoic acid, protocatechuate, pyrocatechol in common for both organisms. 3-Methyl-

TABLE I. GROWTH OF *Alcaligenes* Y42 AND *Acinetobacter* P6 ON VARIOUS AROMATIC COMPOUNDS

The cultivation was continued at least four weeks for poor substrates, and the culture fluid was observed under microscope to make sure the organisms.

Compound	*Alcaligenes* Y42	*Acinetobacter* P6
Biphenyl	++++	+++
4-Chlorobiphenyl	++	+++
4-Methylbiphenyl	++	+++
Benzoic acid	++	++
Monochlorobenzoic acids (2-, 3-, 4-)	–	–
Dichlorobenzoic acid (2, 3-, 2, 4-, 2, 5-)	–	–
2, 3-Dihydroxybenzoic acid	–	–
2, 4-Dihydroxybenzoic acid	–	–
Protocatechuate (3, 4-dihydroxy-benzoic acid)	+	++
2-Phenylphenol	–	–
4-Terphenyl	–	–
2, 2′-Biphenol	–	–
4, 4′-Biphenol	–	–
Diphenylamine	–	–
Diphenylmethane	–	–
Benzhydrol	–	–
Benzophenone	–	–
4, 4′-Dichloro-benzophenone	–	–
2-Chloro-4-phenylphenol	–	–

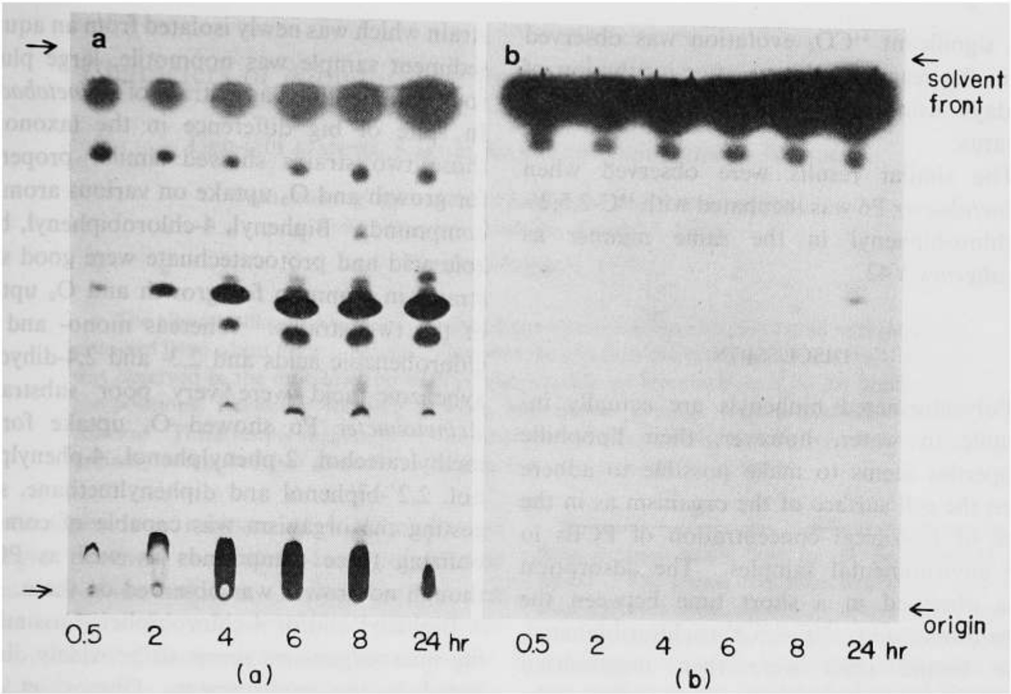

FIG. 2. Representation of the Autoradiograms of the Supernatant Extracts (a) and the Cell 80% Ethanol Extracts (b) as Separated on a Silica Gel TLC.

The TLC plates were developed by using benzene–dioxane–acetic acid (90: 20: 4).

TABLE IV. FRACTIONATED EXTRACTION OF METABOLITES FROM ^{14}C-2, 5, 2′-TRICHLOROBIPHENYL

^{14}C-2, 5, 2′-Used (μg)	Supernatant cpm (%)[a]	Cell extract cpm (%)[a]		Total (%)	Recovery %[b]
		80% ethanol	20% ethanol		
2.6	105279 (88.3)	13498 (11.3)	386 (0.3)	119163 (100)	76.9
12.6	102281 (82.8)	20820 (16.9)	388 (0.3)	123489 (100)	79.7
102.6	91818 (79.5)	23340 (20.2)	266 (0.2)	115424 (100)	74.5
502.6	24030 (24.3)	74466 (75.4)	266 (0.3)	98762 (100)	63.7

[a] Percentage for total cpm recovered.

[b] Percentage recovery for originally used 0.1 μCi.

genes Y42 were incubated for 48 hr. The incubation mixture was fractionated as described in MATERIALS and METHODS, and the radioactivity was measured for each extract (Table IV). When 2,6, 12.6, and 102.6 μg of ^{14}C-2,5,2′-trichlorobiphenyl were used, the radioactivity was mostly measured in the supernatant layer, and the remaining were found in 80% ethanol extract of the cells. However only 24.3%, when 502.6 μg was used, was measured in the supernatant, the remaining radioactivity was in the cell 80% ethanol extract. No significant radioactivity was measured in the cell 20% ethanol extracts in all cases. ^{14}C-Compounds of 80% ethanol cell extracts were mostly detected as unchanged 2,5,2′-trichlorobiphenyl on thin layer chromatography and subsequent autoradiography.