

## 1076b: SP4-Z MEDIUM

This recipe contains strain-specific modifications for *Acholeplasma laidlawii* DSM 23060 \*

Final pH: 7.4

Final volume: 200 ml

1. The difference between this medium and "normal" SP<sub>4</sub> medium (Medium 1076) is that the yeast extract was left out and DNA was added instead. Since most mycoplasmas use either glucose or arginine as their energy source one is added to the media. Do not combine both in one medium, arginine can inhibit the growth of some glucose utilizers (unknown reason).

|                         |        |    |
|-------------------------|--------|----|
| PPLO broth              | 0.70   | g  |
| Tryptone                | 2.00   | g  |
| Bacto peptone (BD)      | 1.00   | g  |
| Distilled water         | 130.00 | ml |
| DNA (fish sperm; SERVA) | 0.04   | g  |

2. Adjust pH to 7.4; autoclave at 121°C for 15 minutes.

3. Add aseptically:

|                                       |       |    |
|---------------------------------------|-------|----|
| CMRL 1066 (10x concentrated; GIBCO)   | 10.00 | ml |
| Yeast extract (BD; 2%, autoclaved)    | 20.00 | ml |
| Fetal bovine serum (heat-inactivated) | 17.00 | ml |
| Swine serum (heat-inactivated)        | 17.00 | ml |

4. As energy source add either:

|   |      |    |
|---|------|----|
| Glucose <b>or</b> L-Arginine x HCl (50 % aqueous) | 2.00 | ml |
|---|------|----|

5. For Ureaplasma add instead of Glucose or Arginine:

|                        |      |   |
|------------------------|------|---|
| Urea                   | 0.40 | g |
| Agar, for solid medium | 2.00 | g |

6. For agar plates use only 126 ml of distilled water and add 2 g of agar. Medium containing agar should be placed at 55°C before adding supplements.

\* Plus glucose; plus 5% CO<sub>2</sub>