Short Communication

WINOGRADSKY'S SALTS SOLUTION AS A DILUTING MEDIUM FOR PLATE COUNT OF OLIGOTROPHIC BACTERIA IN SOIL¹

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JENSEN (1) has pointed out that the composition of a diluting medium for plate count of soil bacteria is of importance and that a salt solution, known as Winogradsky's salt solution, gives a higher count when used as a diluting medium tap water or other salt solutions. In this work we investigated whether Winogradsky's salt solution is also effective or otherwise for plate count of oligotrophic bacteria in soil; the count was obtained as the difference between counts on the full strength nutrient broth (NB) and its 100-fold dilution (DNB) (2).

One gram of Kumanodou loam soil (3) or Sławinek pseudopodzolic soil (4) was dispersed in 20 ml of heat-sterilized water by sonic oscillation (3). The soil suspension was, after being filled up to 100 ml by the same medium, diluted further with heat sterilized water or a modification of Winogradsky's salt solution which was sterilized by passing it through a filter. The latter medium contained 3.8 g of K_2HPO_4 , 1.2 g of KH_2PO_4 , 5.1 g of $MgSO_4 \cdot 7H_2O$, 2.5 g of NaCl, 0.05 g each of $Fe_2(SO_4)_3 \cdot nH_2O$ and $Mn_2(SO_4)_3 \cdot 4H_2O$ in 1,000 ml of deionized water. The method of plate count and the compositions of NB and DNB have been described previously (5). NB was solidified with 1 % of Wako 1st grade agar and DNB 1 % of Agar Purified (Oxoid) (5).

Table 1 shows that the soil suspension with the salt solution gave higher counts on both NB and DNB media than with tap water, and that also the effect varied during the interval preparation of the dilution and plating out. It is notable that, although DNB organisms are sensitive to potassium or sodium salts (2, 6), their counts with the salt solution were higher than with tap water. These results may be closely related to the physiological state of oligotrophic bacteria in soil, and further study should be made to elucidate the mechanism of the effect.

¹ Abbreviations used in this text are as follows: NB, the full strength of nutrient broth; DNB, a 100-fold dilution of nutrient broth.

Table 1. Plate counts of soil bacteria suspended in tap water and Winogradsky's salt solution.

Diluent	Medium	Kumanodou loam soil ^a		Sławinek pseudopodzolic soil ^b	
		Interval between preparation of dilution and plating		Interval between preparation of dilution and plating	
		30 min	1 hr	30 min	1 hr
Tap water	NB	113±23	72.4±7.0	50.8±6.4	49.3±6.3
	DNB	322±25	nd ^c	174.4±15.2	140.8±33.0
	Count of DNB organisms	209	nd	123.6	91.5
Winogradsky's salt solution	NB	152±20	154±19	58.5±3.4	47.8±8.8
	DNB	556±73	580±55	215.2±16.8	268.3±22.3
	Count of DNB organisms	404	426	156.7	220.5

^a Soil suspension used was 10⁶-fold dilution.

^b Soil suspension used was 10⁵-fold dilution.

^c Count was not determined.

REFERENCES

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