EFFECT OF STORAGE TEMPERATURE ON RHIZOBIUM MELILOTI SURVIVAL IN PEAT- AND CLAY-BASE INOCULANTS

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If a legume inoculant is to be an effective product for agriculture it must contain a high population of the appropriate type and strain of rhizobia specific to the legume being planted. Another very important characteristic of an inoculant is the 'survival' of the rhizobia bacteria once the inoculant has left the manufacturer. The present inoculant distribution and marketing systems and the uncertainty of cropping choices necessitate that an inoculant must have a long 'shelf life'. At farm supply dealerships and on farms legume inoculants are usually stored under conditions that are far from the ideal storage conditions available at the manufacturers plant. In the country side inoculants are frequently placed into uninsulated steel sheds in which temperatures can fluctuate widely and can rise far above the constant low temperatures that are considered optimal for maintenance of live bacterial cultures.

To examine the effect of storage temperatures on the survival of rhizobia in a peat- and in a clay-base inoculant, a laboratory storage simulation study was conducted at three different temperatures: -23 C. (freezer), 4 C. (refrigerator) and ambient room temperature (ranging from 21 to 33 C.). Two commercially available products, one peat-based (i.e., Nitragin Co., Western "A" culture containing strain NRG-185) and one clay-based (i.e., Kalo Inc. 'Dormal') alfalfa inoculant, received during the 1987 regulatory Legume Inoculant Testing Program, were used for this study. These powder-type inoculants were divided into a series of 10g subsamples, sealed in polyethylene bags and placed under the three different storage temperatures. Four replicate subsamples of each inoculant and from each storage temperature were analyzed at 2 weeks and at 3, 6, 9, 12, 15 and 20 months of storage. Alfalfa seedlings, cv. 'Algonquin', were used as host plants to determine the number of viable Rh. meliloti cells by means of the modified standard MPN plant infection technique.

The rhizobial population in each of the two inoculant products did not decrease significantly at any of the three storage temperatures tested (Table 1). This remarkably high survival of the Rh. meliloti cells must be attributed to the high initial population level and the high quality of the carrier materials in the two commercial inoculants. Although the two inoculants differed significantly with regard to their initial pH and moisture content, which was 6.8 and 12% in the peat-based and 8.5 and 3% in the clay-based product, these properties remained unchanged in each carrier material throughout the 20 months storage period.

The initial, 2-week and every three month interval MPN count for up to 20 months at storage showed that the viable population remained rather constant around a level of $1x10^9$ and $1x10^8$ rhizobia/g inoculant for the peat-based and the clay-based product, respectively. When the MPN data for each inoculant were averaged across all three storage temperatures and the means were plotted

the resultant graph indicated that both carrier materials were equally effective in sustaining the rhizobial population at its initial level during prolonged storage (Figure 1).

TABLE 1. Survival of Rhizobium in subsamples of peat- and clay-base inoculants during storage.

Type:	peat	clay	peat	clay	peat	clay
Temperature	--- RT ---		--- 4 C. ---		-- -23 C. --	
Storage period:	----------		*Rh. meliloti*/g (Log 10)		----------	
0*			9.3	8.0		
1/2 month	9.2	8.4	9.2	8.3	9.2	8.2
3 months	9.2	7.5	9.2	8.1	9.1	7.7
6 months	8.8	7.5	9.0	8.0	9.1	7.8
9 months	9.2	8.1	9.3	8.1	9.2	8.2
12 months	9.0	7.6	9.0	8.2	9.0	8.3
15 months	8.8	7.5	8.9	8.2	8.8	8.2
20 months	9.1	7.7	9.0	7.9	9.2	8.0

* Initial count on the inoculant previously stored at 4 C.
** All values within each column are not significantly different at the 95% confidence level.
RT Room temperature: 21-33 C.

CONCLUSIONS

A high quality peat- or clay-base inoculant can be effectively stored at ambient room temperature (ranging from 21 to 33 C.), 4 C. (refrigerator), or -23 C. (freezer), for up to 20 months without significant reduction in rhizobial survival.

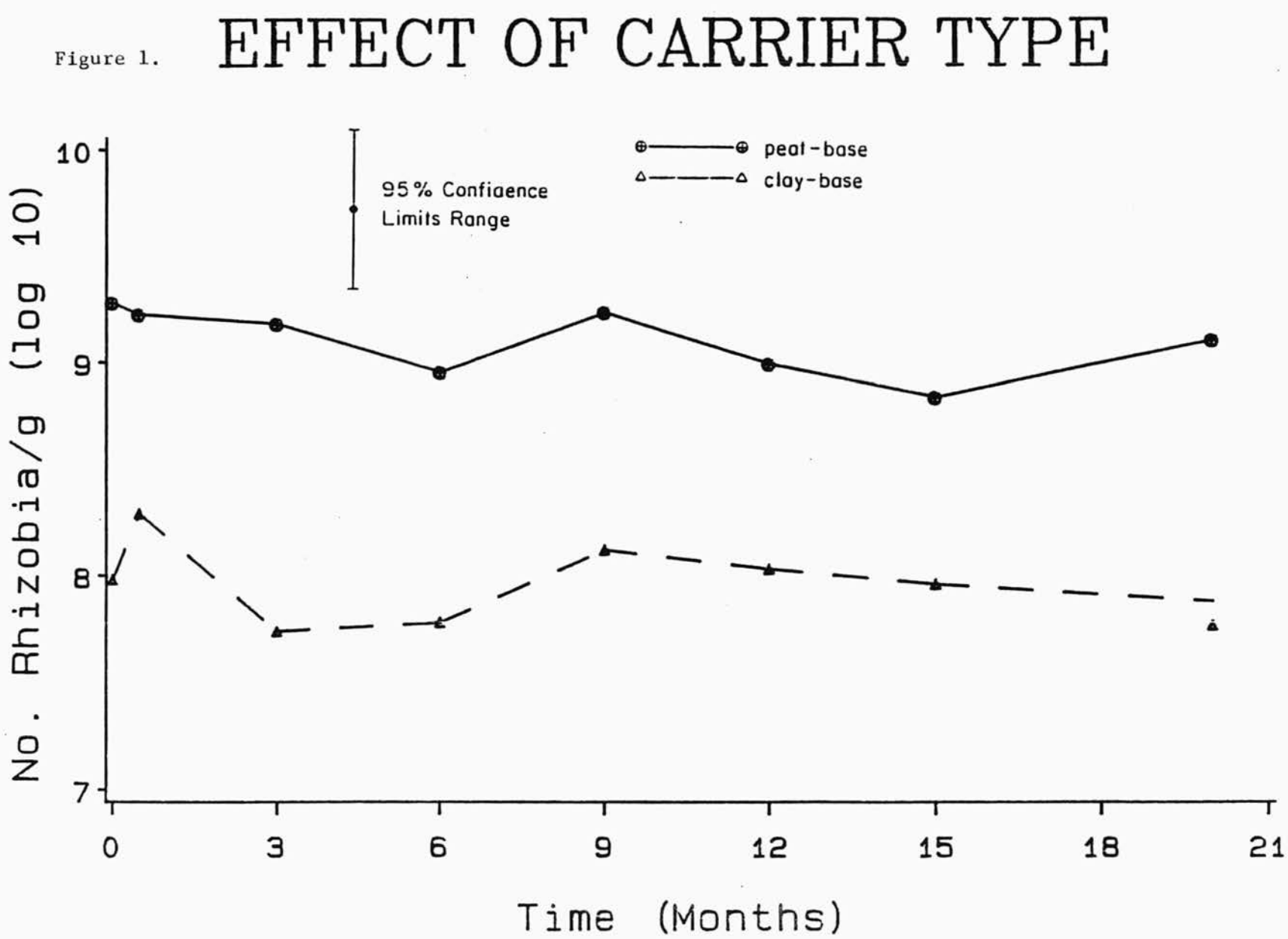
Since not all commmercially available peat- or clay-base inoculants are of the same high quality as those used in this experiment, they may not display the same level of survival in storage.

Averaging the storage data across all three temperatures for each carrier type suggests that both carriers can sustain a high degree of rhizobial longevity.

All samples tested, at any storage period, met the Fertilizers Act's minimum requirement for rhizobial content and would thus be rated as satisfactory products.

Since inoculant quality is not readily known to a legume grower, and we have not tested any 'marginal' quality inoculant products during storage we still recommend storage at low temperatures if possible. Until all commercially available inoculants are of consistant high quality, the 4 C. storage will continue to be the recommended storage condition.

Figure 1.


EFFECT OF CARRIER TYPE
peat-base
clay-base
95% Confidence
Limits Range
No. Rhizobia/g (log 10)
10
9
8
7
0
3
6
9
12
15
18
21
Time (Months)