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Vitamins for Algal Chemostats

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This a protocol to make vitamins for algal chemostats that are used to feed Daphnia and Ceriodaphnia in the Duffy Lab.

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VITAMINS – CONTENTS

A	B	C
VITAMIN	mg/2L	Sigma Cat#
1. Biotin	10	B4501
2. Thiamine	200	T4625
3. Pyridoxine	200	P9755
4. Pyridoxamine	6	P9380
5. Calcium pantothenate	500	P2250
6. B 12	2	V2876
7. Nicotinic Acid	100	N4126
8. Nicotinamide	100	N3376
9. Folic Acid	40	F7876
10. Riboflavin	60	R4500
11. Inositol	180	I5125

Since algae do not supply all the requirements for zooplankton culture, a mixture of eleven vitamins is added to the algal culture while it is in exponential growth phase.
To make the mixture, compounds are weighed on the analytical balance and combined in 2L volumetric flask containing approximately 1800 ml of MilliQ water. Cover the flask with aluminum foil or a box and allow to mix on a stir plate until the vitamins have dissolved into a clear yellow color. Fill to 2L with MilliQ water. A minimum volume of 2L is used in order to have a weighable amount of pyridoxamine and B 12.
The solution is then dispensed into dated 6 oz. Whirl paks and frozen.
As needed, vitamins are thawed and poured into sterile, microcentrifuge tubes. The tubes are then refrozen and stored in the dark since vitamins are sensitive to light and heat.
The mixture can kept frozen for more than a year and at 4-6 C for several weeks.
1-2 ml of vitamin mixture is normally added for each liter of media solution. Vitamins are added to the algae by withdrawing the desired amount using a syringe and injecting it into the silicone tubing while adding media to the algal culture.