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# MELiSSA – Adaptation for Space

ESA contract 15671/01/NL/ND

## TECHNICAL NOTE 72.7.4

### Trade-off of solid-liquid separation and desalination technologies and concept of breadboard

Version: 2  
Issue: 1

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18/03/2003

## DOCUMENT CHANGE LOG

<b>Version</b>	<b>Issue</b>	<b>Date</b>	<b>Observation</b>
1	0	31/12/2002	Draft
2	1	18/03/2003	Final

## DISTRIBUTION LIST

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# 1. Introduction

In the MELiSSA loop, the liquid solid separation of *Arthrospira platensis* in Compartment IVa is probably the most critical and challenging one. In order to be able to design and construct a breadboard for harvesting the *Arthrospira* cells and washing the harvested biomass, different technologies have been tested in the past months to evaluate their potential as either harvesting or desalination technique. The candidate concentration techniques were ultrasonic separation, membrane filtration and centrifugation; the candidate desalination techniques were reverse osmosis and electrodialysis. The results of the experimental work have been presented and discussed in Technical Note 72.7.3.

In the present Technical Note, a trade-off will be made among the alternative harvesting and desalination technologies. This will allow us to present a general concept for breadboard design.

## 2. Trade-off of technologies

The trade-off between alternative technologies will be performed using predefined criteria for a given set of boundary conditions. Because these are evidently different for concentration technologies and desalination technologies, the evaluation will be presented separately.

### 2.1 Trade-off of harvesting technologies

The liquid-solid separation systems considered for the trade-off, were those, which had previously been selected and tested: ultrasonic separation, membrane filtration and centrifugation.

The following assumptions were made for the trade-off:

- Harvest of 100 l *Arthrospira* suspension per day
- The harvested suspension of *Arthrospira* has a concentration of 1 g/l
- The cells need to be concentrated by a factor of at least 10 to a final concentration of between 10 and 20 g/l

The criteria, which were considered essential for evaluation of a particular liquid-solid separation technology, were the following:

- Separation efficiency: defined as the difference in biomass concentration between feed solution and clarified water, divided by the biomass concentration in the feed
- Breakthrough of cells: indicating whether or not a risk for breakthrough of cells exists. The ultrasonic separation system for example, does not provide an actual filter for the cells but retains them by an invisible mesh. Although the separation efficiency at optimal operation amounts to e.g. 95%, at some point, the cell concentration in the chamber will become so high that the ultrasonic forces in the chamber cannot withhold the cells and breakthrough will occur.
- Energy requirement: calculated in kWh/m<sup>3</sup> of harvested biomass. To avoid excessive energy needs when considering small volumes of treated suspension, the total volume was set to 100 l/d as indicated in the assumptions. Energy requirements contribute to equivalent system mass and should be as low as possible.

- Biomass integrity: because the harvested cells are to be recovered as edible biomass and because the nutritional quality is related to their integrity, the harvesting system should ideally not cause any damage to the cells.
- Biomass recovery: the intended use of the harvested biomass as food also implies that the concentrated cells can be recovered from the liquid-solid separation system. In membrane filtration for example, part of the biomass will stick to the membrane. It can be removed by chemical cleaning, but will no longer be suitable for consumption.
- Water recovery: this parameter is important because the salt-rich Zarrouk medium, in which the cells are grown, will be recovered for two purposes. First of all, the salts should be recycled to compartment IVa to avoid the need for a high external supply. Secondly, it is the intention to desalinate the Zarrouk medium. The desalinated water will be used to wash the harvested biomass and remove the salty taste. For both reasons the water recovery should be as high as possible. A 100% recovery is however not realistic. To remove the harvested cells from the liquid-solid separation system, they will have to be resuspended in water. Alternatively, the system only concentrated them to a reduced final volume.
- Consumables: particularly for long-term space missions the use of chemicals needs to be limited and also the type of chemicals allowed is strictly regulated. Therefore, the necessity of consumables is a drawback for a particular liquid-solid separation system.
- Mass: this is one of the limiting factors for space flights. This parameter contributes to the equivalent system mass and needs to be as low as possible.
- Safety issues: these include the presence of rotating parts, operation at high pressures, etc. as the case of centrifugation.
- Potential for improvement in space: this criterion evaluates whether the possibility exists to adapt a terrestrial technology for application in space. For example, the dependence of a technology on gravity can in some cases be overcome by creating artificial gravity forces.

For each criterion different classes were defined and given a score.

- Separation efficiency: 0% = score 0, 50% = score 1, 95% = score 2, 100% = score 3
- Breakthrough of cells: yes = score 0, no = score 1
- Energy requirements:  $> 25 \text{ kWh/m}^3$  = score 1,  $< 25 \text{ kWh/m}^3$  = score 2
- Biomass integrity: no = score 0, 50% = score 1, 90% = score 2, 100% = score 3
- Biomass recovery: no = score 0, 50% = score 1, 90% = score 2, 100% = score 3
- Water recovery: 90% = score 1, 95% = score 2
- Consumables: yes = score 0, no = score 1
- Mass: = score 0, = score 1, = score 2, = score 3
- Safety issues: not adapted = score 0, adapted = score 1
- Potential of improvement for space: no: score 0, yes = score 1

Finally, each criterion was assigned a weight factor, which is only valid for selection of harvesting technologies in the framework of the present project. Those criteria, which were considered to be crucial for the harvest of *Arthrospira* and its subsequent use as food (for a terrestrial demonstration), were given a weight factor of 100. These are separation efficiency, biomass integrity and biomass recovery. The other criteria were assigned a weight factor of 50.

This gives the results presented in Table 1. Based on the weight factors and the scores, the technique with the highest total score is preferred to the alternatives with lower scores.

Table 1. Trade-off for selected liquid-solid separation technologies under the assumption that 100 l of *Arthrospira* suspension is harvested per day at a concentration of 1 g/l and is concentrated to between 10 and 20 g/l. Technology A: ultrasonic separation, B: ultrafiltration, C: A + B, D: centrifugation.

	CRITERIA	UNITS	WEIGHT	A	B	C	D
1	Separation efficiency	%	100	200	300	300	200
2	Breakthrough of cells		50	0	50	50	0
3	Energy requirements	KWh/m <sup>3</sup>	50	100	50-100	50	50
4	Biomass integrity	%	100	300	100-200	300	200
5	Biomass recovery	%	100	200	200	300	200
6	Water recovery	%	50	50	50	50	100
7	Consumables		50	50	0	0	50
8	Mass	Kg/m <sup>3</sup>	50	0	50	0	0
9	Safety issues		50	50	50	50	0
10	Potential of improvement for space		50	50	50	50	0
	<b>Total score</b>			<b>1000</b>	<b>900-1050</b>	<b>1150</b>	<b>800</b>

1	2	3	4	5
0% = 0	YES = 0	> 25 kWh/m <sup>3</sup> = 1	NO = 0	NO = 0
~50% = 1	NO = 1	< 25 kWh/m <sup>3</sup> = 2	~ 50% = 1	~ 50% = 1
~95% = 2			~ 90% = 2	~ 90% = 2
100% = 3			100% = 3	100% = 3
6	7	8	9	10
90% = 1	YES = 0	> 500 kg/m <sup>3</sup> = 0	Not adapted = 0	NO = 0
95% = 2	NO = 1	100 kg/m <sup>3</sup> = 1	Adapted = 1	YES = 1
		50 kg/m <sup>3</sup> = 2		
		10 kg/m <sup>3</sup> = 3		

As discussed in Technical Note 72.7.3, ultrasonic separation can achieve a separation efficiency of 95% and higher. The risk of breakthrough exists because no physical barrier for cell retention is present. Therefore, biomass recovery is lower than 100%. Energy consumption has been calculated to be around 8 kWh/m<sup>3</sup>. Biomass integrity is not affected by this technique. Water recovery will be around 90% for a tenfold increase in cell concentration. Consumables are not required for proper functioning of the system. Mass was calculated to be 2100 kg/m<sup>3</sup>. The system does contain pumps but does not operate at high temperature or pressure. Therefore it scores better than centrifugation with respect to safety issues. It also shows potential to be improved for space. Ultrasonic separation now depends to some extent on sedimentation of the cell aggregates. However, gravity influence can be replaced by applying suction.



Ultrafiltration yields 100% separation efficiency since the membrane retains all cells. Energy requirements are estimated to be between 5 and 30 kWh/m<sup>3</sup>. Biomass integrity is affected in the process due to high-speed recirculation of the cells over the membrane. Between 50 and 90% of the cells were found to be damaged in the experimental work. Biomass recovery amounts to 90%. The remainder of the biomass is lost due to adsorption to the membranes. Water recovery is close to 90% at a 10-fold concentration factor. Membrane processes inevitably require consumables e.g. for membrane cleaning. Mass is estimated to be over 100 kg/m<sup>3</sup>. In terms of safety, no specific problems are envisaged because ultrafiltration is performed at fairly low pressures. The process does however show potential for improvement for space.

The experimental work on centrifugation indicated that a separation efficiency of around 95% can be achieved. As for ultrasonic separation, a risk of cell breakthrough exists. Energy requirements have been calculated to amount to 45 kWh/m<sup>3</sup>. Damage to cells has been observed but is limited to around 10%. Because the separation efficiency does not equal 100%, biomass recovery is estimated to be around 90%. Water recovery is close to 95%, since some water volume has to be used to remove the cells from the centrifuge. Consumables are not required. The mass of the system amounts to 1100 kg/m<sup>3</sup>. Centrifuges operate at high rotation speeds and are therefore a problem with respect to safety issues. They also do not show any potential for adaptation to space conditions and restrictions.

Centrifugation and ultrasonic separation have the disadvantage that they do not show a 100% cell separation. Breakthrough of cells has negative implications on the desalination technology to which the clarified water will presumably be fed (see 3). In addition, the clarified water needs to be recycled to Compartment IVa because of its salt content. Preferably, it should be free of dead cells and cell debris to avoid accumulation and toxic effects. For both reasons, centrifugation and ultrasonic separation cannot be used as stand-alone techniques for cell harvesting. Combination with a membrane filtration step has the advantage that a 100% cell removal can be achieved. Moreover, the removal of the majority of the cells in a previous treatment step improves the performance of membrane filtration. The potential of membrane fouling will be lower and higher recoveries can most probably be achieved.

Because centrifugation scores lower than ultrasonic separation in the trade-off and because it specifically does not respond to space requirements, the combination with membrane filtration was only evaluated for the latter technique. As shown in Table 1, the combination of ultrasound and ultrafiltration scores better than ultrasound alone because it prevents the breakthrough of cells and hence achieves a 100% separation efficiency. Energy requirements were estimated to be 50 kWh/m<sup>3</sup> at maximum, assuming that the energy requirements for ultrasound and membrane filtration are similar in the combined set-up as for the separate techniques. Because the major part of the biomass is eliminated from the medium by ultrasonic separation, the overall biomass integrity will be close to 100%, as for ultrasound alone. Likewise, biomass recovery will be higher than for ultrafiltration alone because the amount of cells adsorbed to the membrane will be much lower. Water recovery will supposedly be close to 90%. On the one hand, concentration factors for ultrasonic separation are between 10 and 20. On the other hand, the recovery factor for membrane filtration will probably be higher than for ultrafiltration alone due to the much reduced cell concentration. For the criteria consumables, mass, safety and potential of improvement for space the scores have been explained before.

It is logical that the combination of ultrasound and ultrafiltration scores better than the individual techniques. Still, the differences cannot be considered significant because they do not exceed 20%.

As a conclusion, it can be stated that the scores for ultrasonic separation, centrifugation and ultrafiltration did not differ much. However, centrifugation does not respond to space requirements. Ultrasound and membrane filtration together will probably give the best result in terms of cell separation efficiency, biomass recovery and integrity. In addition, they yield a clarified medium that can directly be fed to a desalination step.

## 2.2 Trade-off of desalination technologies

As candidate desalination technologies, reverse osmosis and electrodialysis have been tested and will be compared.

Both techniques will be compared in worst-case conditions, namely the desalination of Zarrouk medium down to a final salinity of 0.3 g/l. It can be doubted that in the final MELiSSA loop a medium with such a high salt concentration will ever be used. However, since all the experimental work on growth kinetics of *Arthrospira* has been performed in Zarrouk medium, it was also used for the desalination tests. The final salt concentration was defined previously to produce a food product suitable for human consumption. Furthermore it was assumed that desalination would occur batchwise. Most probably, harvesting of the biomass will also occur in batch mode, e.g. once a day. In addition, the biomass needs to be desalted in a stepwise washing procedure, for which the washing water will be generated by repeated desalting of the water volume from the previous washing step. In other words, the same volume of water obtained after cell harvesting, will probably need to be desalted three times in the course of one day. This can be performed most conveniently in a batchwise mode of operation.

The criteria for trade-off are the following:

- Separation efficiency: in this case referring to the difference in salt concentration in feed water and desalted stream
- Energy requirements: these have been calculated per m<sup>3</sup> of permeate or diluate
- Water recovery: this equals the ratio of permeate to feed and depends on the danger for scaling of the membranes for membrane processes. The higher the scaling potential, the lower the recovery that can be achieved without operational problems.
- Salts recovery: refers to the fact that e.g. some salts may be lost by irreversible adherence to membranes
- Consumables: as described in 2.1
- Mass: as described in 2.1
- Safety issues: as described in 2.1
- Potential of improvement for space: as described in 2.1

For each criterion different classes were defined and given a score.

- Separation efficiency: 0% = score 0, 50% = score 1, 95% = score 2, 100% = score 3
- Energy requirements: > 25 kWh/m<sup>3</sup> = score 1, < 25 kWh/m<sup>3</sup> = score 2
- Water recovery: 90% = score 1, 95% = score 2, 100% = score 3
- Salts recovery: no = score 0, 50% = score 1, 90% = score 2, 100% = score 3
- Consumables: yes = score 0, no = score 1
- Mass: > 500 kg/m<sup>3</sup> = score 0, 100 kg/m<sup>3</sup> = score 1, 50 kg/m<sup>3</sup> = score 2, 10 kg/m<sup>3</sup> = score 3
- Safety issues: not adapted = score 0, adapted = score 1

- Potential of improvement for space: no = score 0, yes = score 1

As for the liquid-solid separation technologies, weight factors were assigned to the different criteria depending on their importance in the present project. The criteria separation efficiency, water recovery and salts recovery were given a weight factor of 100, the others 50.

The results are summarized in Table 2. Electrodialysis and reverse osmosis can achieve similar separation efficiencies in batch operation. Energy requirements are fairly similar but can be somewhat higher for electrodialysis when high voltages are applied. The data used here for reverse osmosis differ from those given in Technical Note 72.7.3 in that they are here calculated for batch operation of 4 2.5 inch modules (type SW30-2514) and hence have to take into account a flux decline due to increased salt concentrations in the concentrate. Water recovery was set to 100% for both techniques because the concentrate can be recycled to compartment IVa with recovery of the salts from the Zarrouk medium. The filtrate or diluate is used for cell washing. No salts are lost during the process. Even if water and salts recovery was lower, the performance of both techniques for these criteria would in any case be comparable. Both technologies require the use of chemicals for membrane cleaning and anti-scalants. Particularly the high carbonate concentration in Zarrouk presents a high potential for scaling. Exact weights are not known, but it is expected that the mass for reverse osmosis is higher than for electrodialysis, because it requires a heavy pressure pump and materials to withstand the high operating pressure. The high operating pressure (up to 55 bar) also poses problems in terms of safety.

Table 2. Trade-off for selected desalination technologies under the assumption that Zarrouk medium is desalinated batchwise to a final salinity of 0.3 g/l. Technology A: electrodialysis, B: reverse osmosis.

	<b>CRITERIA</b>	<b>UNITS</b>	<b>WEIGHT</b>	<b>A</b>	<b>B</b>
<b>1</b>	Separation efficiency	%	100	300	300
<b>2</b>	Energy requirements	KWh/m <sup>3</sup>	50	50-100	100
<b>3</b>	Water recovery	%	100	300	300
<b>4</b>	Salts recovery	%	100	300	300
<b>5</b>	Consumables		50	0	0
<b>6</b>	Mass	Kg/m <sup>3</sup>	50	50	0
<b>7</b>	Safety issues		50	50	0
<b>8</b>	Potential of improvement for space		50	50	50
	<b>Total score</b>			<b>1100-1150</b>	<b>1050</b>

<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>
0% = 0	> 25 kWh/m <sup>3</sup> = 1	90% = 1	NO = 0
~50% = 1	< 25 kWh/m <sup>3</sup> = 2	95% = 2	~ 50% = 1
~95% = 2		100% = 3	~ 90% = 2
100% = 3			100% = 3
<b>5</b>	<b>6</b>	<b>7</b>	<b>8</b>
YES = 0	> 500 kg/m <sup>3</sup> = 0	Not adapted = 0	NO = 0
NO = 1	100 kg/m <sup>3</sup> = 1	Adapted = 1	YES = 1
	50 kg/m <sup>3</sup> = 2		
	10 kg/m <sup>3</sup> = 3		

The trade-off table does not show large differences for both techniques. For the final concept preference is given to electro dialysis for the following reasons:

- The final salinity of the filtrate can easily be adjusted to any desired level. It can be expected that during subsequent washing cycles of the harvested cells, water of decreasing salinity will be used. Electro dialysis has the flexibility to provide water of any salinity, whereas a specific reverse osmosis system has not.
- The system generally has a lower potential for scaling and uses less chemicals
- The system is preferable in terms of safety

### 3. Compatibility of selected techniques with microgravity

Of the above-mentioned techniques, only ultrasonic separation is gravity-dependent. In membrane techniques, liquid pumping provides a shearing force across the membranes to reduce fouling and it generates the pressure difference over the membrane, which is the driving force for the permeation process. In centrifugation gravity forces are generated by high-speed rotation. For ultrasonic treatment however, part of the separation process consists of a settling of aggregates. As described in technical note 72.7.3, the cell suspension is circulated from a reactor or tank through a resonance chamber back to the reactor at a given recirculation rate. A second pump operating at about one third of the recirculation rate, drags clarified water (= harvest) out of the chamber at the top. Acoustic forces retain the cells in nodal planes where they form loose clumps. As long as the ultrasonic field is switched on, the clumps are held stationary against the fluid drag in the chamber. However, to prevent clogging of the chamber with cells, the field needs to be switched off at regular time intervals. During that period, the pump in the harvest line is switched off and the aggregates settle due to gravitational forces.

To eliminate the dependency of ultrasonic separation on gravity, two approaches can be envisaged. On the one hand, a suction could be applied on the recirculation line to drag the aggregates back into the reactor when the ultrasonic field is switched off. On the other hand, a prime rate reverse pump can be used in the harvest line. Crognale et al. (2002) found it necessary to use this type of pump for the separation of filamentous fungi. It has the advantage that it automatically reverses the flow direction when the ultrasonic field is switched off. However, attention has to be paid to the fact that the resonance chamber may be completely empty of the cell suspension and part of the clear filtrate in the harvest tube may return into the chamber. Therefore, stop times should be sufficiently short.

Concerning the desalination technologies, both electro dialysis and reverse osmosis are independent of gravity because the driving force for the transfer of water and/or salts is either pressure or a potential difference.

### 4. Concept of breadboard

In this paragraph, a concept of breadboard for harvesting and washing of *Arthrospira* is presented. The boundary conditions are the following:

- *Arthrospira* is grown in Zarrouk medium at concentrations of around 1 g/l. However, the harvesting system must be able to cope with variations in growth rate, flow, cell concentration,..etc.
- The harvesting mode is preferentially continuous. Or, if operated in batch mode, the harvesting system should not interfere with the continuous operation of either *Arthrospira* compartment IVa as such or the MELiSSA loop as a whole.

- The nutritional quality of *Arthrospira* quickly deteriorates upon storage. It is assumed that conservation at 4°C during one day is the maximum.
- The minimal harvest volume is 5-10 l.
- *Arthrospira* needs to be concentrated to a final concentration of between 10 and 20 g/l
- The harvested cells need to be washed to a final salt concentration of 0.3 g/l. Since the original salt concentration in Zarrouk medium is well above 20 g/l, several washing cycles at tenfold dilution need to be performed.
- The algae suspension collected from compartment IVa over a period of one day should be harvested and washed over one working day (8 hours) to prevent deterioration of nutritional quality and for practical reasons. If after the concentration step e.g. 3 washing cycles have to be performed, the total duration of one cycle should not exceed 2 h. This time restriction will determine the size of the breadboard.
- The ultrasonic separation device proposed in the actual breadboard is not adequate for harvesting high volume of the alga. An upgraded version of the Ultrasonic separation apparatus type Applisens is possible to handle the volumes required in the MELISSA loop.
- During harvesting the cells will be concentrated and clear water generated. This water can be desalinated and reused in the different washing steps. Desalination is however not considered to be a critical step in the breadboard demonstration because another fresh water source can be used for washing. In fact, large amounts of high quality water are available in the MELiSSA loop from the higher plant compartment.
- The harvesting of *Arthrospira* should be as simple as possible, in terms of numbers of pumps, tanks, etc. to be used.

Following the trade-off on harvesting and desalination technologies, liquid-solid separation will be performed by ultrasonic separation and ultrafiltration, desalination of the clarified water by electrodialysis. The overall schematic is presented in Figure 1.

Effluent from compartment IVa is collected in a so-called concentration tank, preferably at 4°C, over a period of one day. The actual harvest process is initiated by concentrating the algae suspension 10-20 fold by ultrasonic separation. This process unit requires two pumps, one for recirculation of the suspension through the resonance chamber and one to collect the harvest at the top of the chamber. Because the separation efficiency of ultrasound is around 95%, the outlet stream is further clarified by ultrafiltration. The permeate is either sent to the electrodialysis unit or recycled to compartment IVa to reuse the salts. The concentrate contains the remaining cells and is combined with the largest fraction of harvested cells in the concentration tank. In order to provide a sufficiently high cross flow velocity along the membranes, an additional pump is required. When needed at some point, part of the concentrate can be wasted. In the electrodialysis unit, the feed is split into a filtrate which is returned to the concentration tank to initiate the first washing cycle and into a concentrate which is recycled to compartment IVa to reuse the salts. A drain is provided as well.

A washing cycle essentially consists of the same steps as those described above for the first concentration step. When the desired final salinity of the algae suspension is achieved, the concentrated cell suspension will be drained and further processed for human consumption. At that point, the next batch of algae can be harvested and washed.

For several units in the treatment train, a batchwise approach is the best way to go ahead. In the ultrasonic step, a 10-20-fold cell concentration is desired, which can only be achieved in batch mode. In electrodialysis, a continuous operation would imply that the desired final salinity of 0.3 g/l in the filtrate

has to be present at the very start of the test. In addition, the degree of automation provided for the breadboard will probably not be high enough to operate all units simultaneously. Therefore, at this stage, preference is given to a sequence of operations. This explains why batch tanks are as yet provided in between the process units. The detailed design of the breadboard will focus on a potential reduction in the number of tanks and pumps.

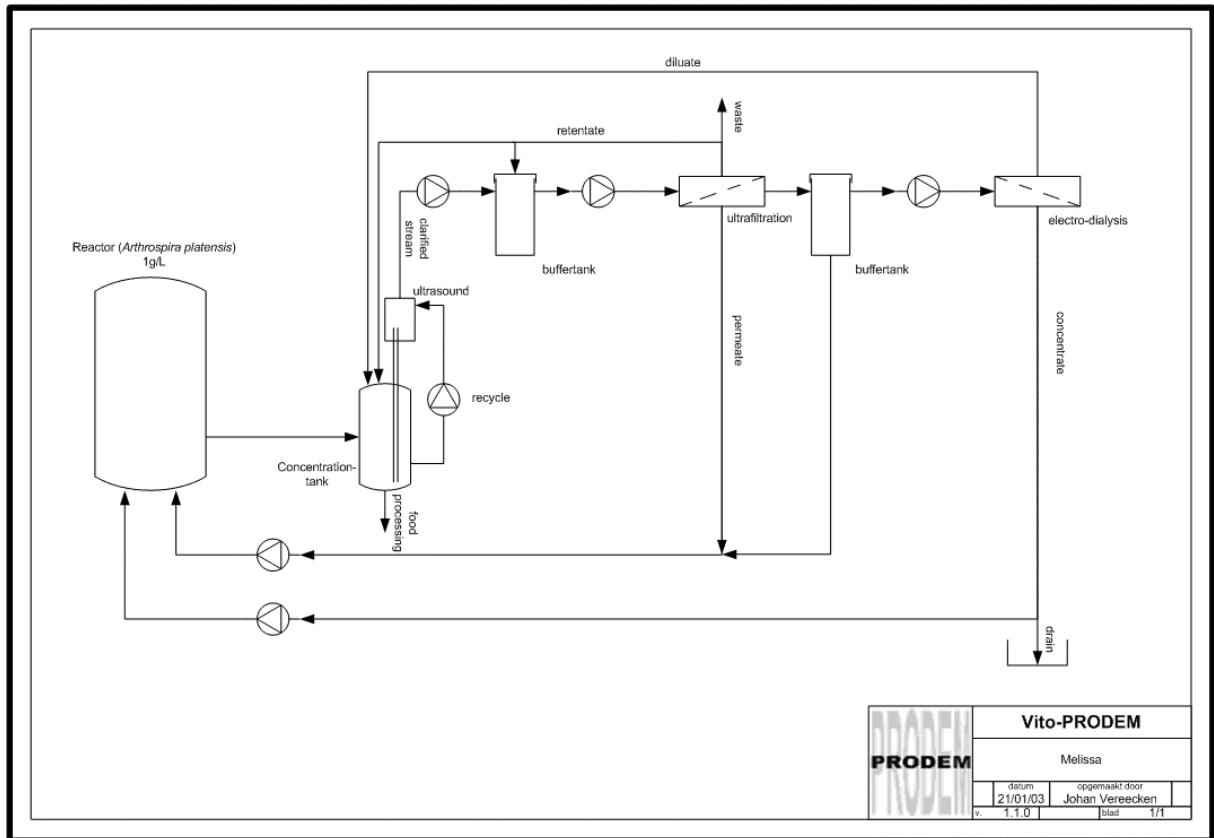


Figure 1. Schematic representation of the breadboard for liquid-solid separation and washing of *Arthrospira* suspensions generated by Compartment IVa.

## 5. Reference

Crognale, S., Federici, F., Petruccioli, M. 2002. Enhanced separation of filamentous fungi by ultrasonic field: possible usage in repeated batch processes. *Journal of Biotechnology* 97: 191-197