The problems with the digestion of foliage samples

in Central Forest Laboratory Zvolen

Author: Ing. Dana Krupova

Národné lesnícke centrum
T. G. Masaryka 22, 960 92 Zvolen (SK)
tel.: +421/(0)45/5320316, fax: +421/(0)45/5314192
e-mail: nlc@nlcsk.org

The problems with the digestion of foliage

1. The digestion in 20 years old microwave oven MDS
2. The using new Microwave oven Mars - X press
3. The evaluation of variability and the recovery of different digestion program choosing the best
4. Using this program make the comparison of CV and Recovery for the same elements of different samples
5. The cooperation Comenius University
6. Next problems

Národné lesnícke centrum
T. G. Masaryka 22, 960 92 Zvolen (SK)
tel.: +421/(0)45/5320316, fax: +421/(0)45/5314192
e-mail: nlc@nlcsk.org

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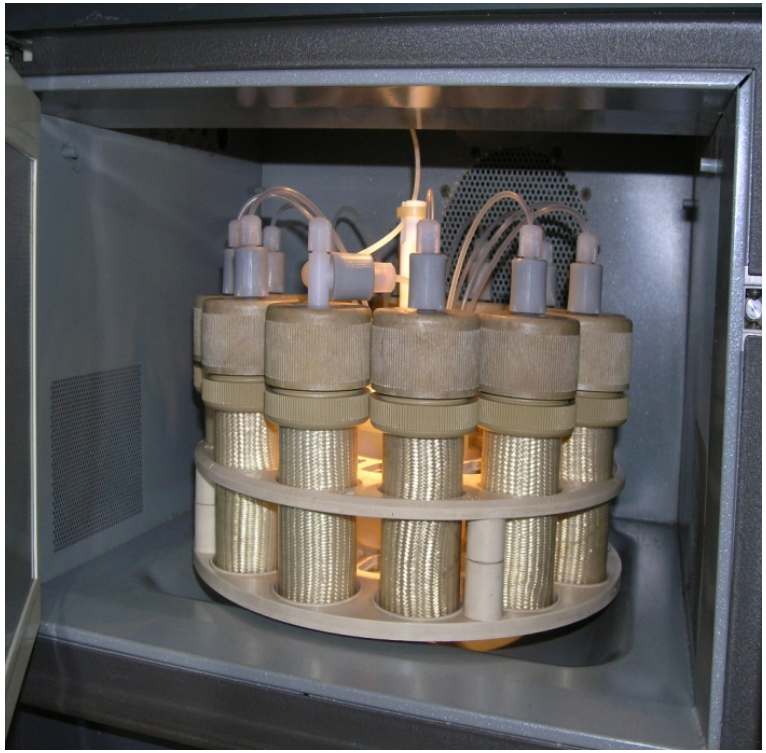

OVEN MDS-2000

- We decided to replace 20 years old microwave oven
- It had pressure control of digestion
- Teflon 150 ml vessel
- Rotary plate for twelve samples

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T. G. Masaryka 22, 960 92 Zvolen (SK)
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Method of MDS digestion

- Weight 0.5 g grinned homogenised sample with the precision 0.0005 g
- Add 5 ml nitric acid and 2ml deionised water
- Wait 30 minutes
- Four stages of digestion in oven each one lasts10 minutes
- High pressure 170 PSI
- Digested sample pour into volumetric 50 ml flask quantitatively
- Fill up to 50 ml deionised water
- Filtrate to the PE vessels of 100 ml

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T. G. Masaryka 22, 960 92 Zvolen (SK)
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New microwave oven Mars X-Press

- Temperature control of digestion
- Maximum power 1200 W
- 40 places for digestion
- Teflon vessels of 55 ml volume
- Till now we use only programs for 20 vessels
- We help ourselves by application notes for organic samples
- Follow variability and recovery

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T. G. Masaryka 22, 960 92 Zvolen (SK)
tel.: +421/(0)45/5320316, fax: +421/(0)45/5314192
e-mail: nlc@nlcsk.org

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New microwave oven Mars X-Press

- At first we use program recommended by technician
- Power 1200 W
- 80 % of power for 20 vessels
- RAMP 20 minutes
- Temperature **190 °C**
- HOLD 15 minutes
- Cooling 30 minutes
- **Recovery** most of the samples was only between **8O – 9O %**

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T. G. Masaryka 22, 960 92 Zvolen (SK)
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Results of digestion

- **The variability** of determination was too high about 10 -15 %
- The technician recommended us to use instrument for tight closing the vessel **CAPPING STATION**
- Then variability improves
- We try next program to improve recovery of elements

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e-mail: nlc@nlcsk.org

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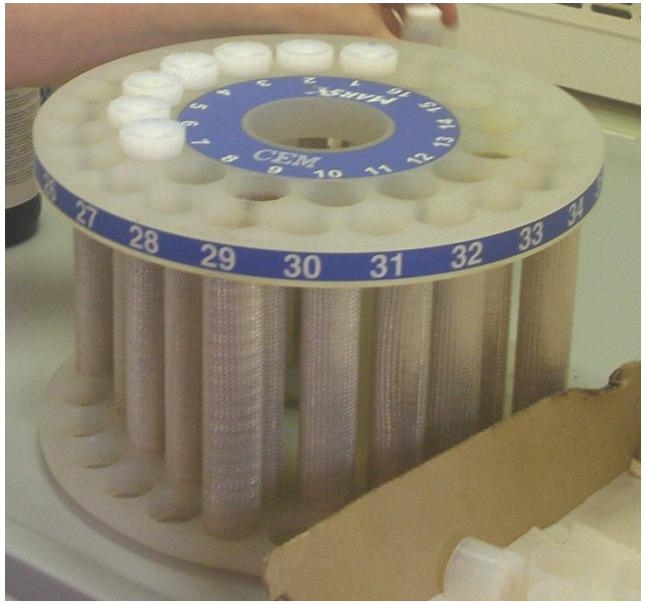

Digestion programme

- According the literature we decided to decrease temperature to 17O C and increase time of Ramp
- **Programme I**
- Power 1200 W
- 75 % of power
- RAMP 30 minutes
- Temperature 170 C
- HOLD 10 minutes for 20 vessels
- Cooling 30 minutes

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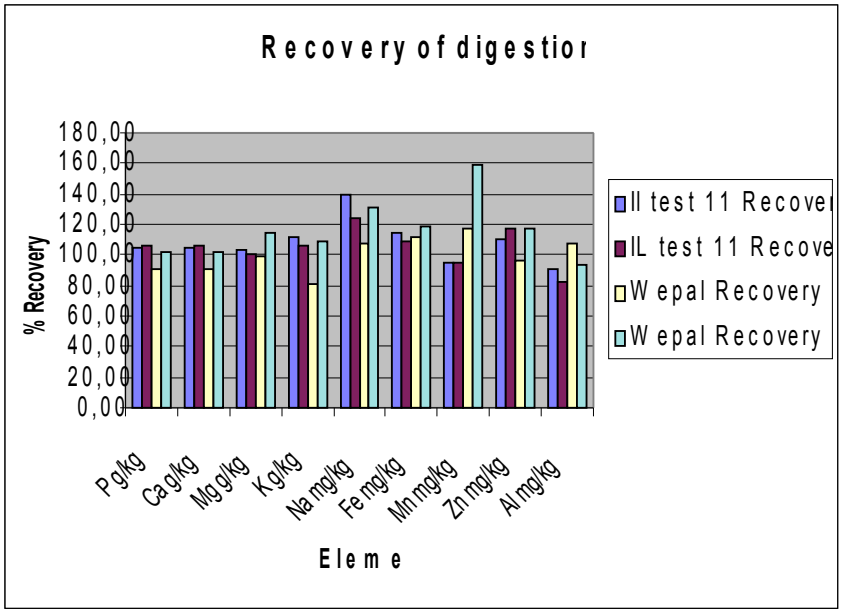
		IL-test _11		Wepal	
		Recovery %		Recovery %	
	Element	MDS	Mars	MDS	Mars
1	**P g/kg**	104,75	106,03	90,55	102,19
2	**Ca g/kg**	104,75	106,03	90,55	102,19
3	**Mg g/kg**	103,03	100,78	99,76	114,14
4	**K g/kg**	112,10	106,73	81,54	108,47
5	**Na mg/kg**	139,57	123,81	107,64	**131,64**
6	**Fe mg/kg**	113,90	109,44	112,13	119,18
7	**Mn mg/kg**	95,07	95,21	117,28	**158,68**
8	**Zn mg/kg**	109,64	117,78	95,66	117,46
9	**Al mg/kg**	90,54	82,61	108,04	93,26
	Average	**108,15**	**105,38**	**100,35**	**116,36**



Národné lesnícke centrum
T. G. Masaryka 22, 960 92 Zvolen (SK)
tel.: +421/(0)45/5320316, fax: +421/(0)45/5314192

The problems with the digestion of foliage

Remedy of bad results

- Recovery of some elements was too high
- We try the other program of digestion
- Increase temperature
- Using longer time for Ramp
- Longer time for HOLD
- Change % of power
- Using double step digestion

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T. G. Masaryka 22, 960 92 Zvolen (SK)
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Program of digestion

Heating Program : Ramp to temperature Control

	stage	Max Power	% Power	RAMP (min)	temperature	Hold (min)
Program I	1.	1200	75	30	170	15
Program II	1.	1200	75	30	190	15
Program III	1.	1200	100	30	190	15
Program IV	1.	1200	65	10	130	10
	2	1200	85	10	190	15

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CV % and Recovery of different digestion methods using sample Wepal IPE 100

CV %	sample	1	2	3	4	5	6	7	8	9	10	average
Recovery	WEPAL 100	PT	CaT	MgT	KT	NaT	FeT	MnT	ZnT	Al	BT	recovery
Method	reference	38,4	55,4	20	261	33	5,12	0,80	0,33	7,05	0,095	elements
digestion	value	g/kg	g/kg	g/kg	g/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	
program	170 C	3,4	9,6	15,4	20,3	21,2	16,6	19,0	19,1	12,8	23,4	16,09
I.	75%	84,6	88,3	96,4	104	113	91,0	108	95,0	39,7	66,2	88,6
program	190 C	2,6	6,1	11,2	19,8	20,1	18,9	22,1	6,0	15,8	15,9	13,87
II.	75%	87,7	88,5	97,4	109,2	119,9	108,7	113,6	88,2	57,8	76,0	94,7
program	190 C	4,9	3,2	5,8	20,2	21,6	11,1	4,3	6,4	10,3	5,9	9,37
III.	100%	87,0	88,6	94,3	98,4	106,9	91,0	94,6	77,8	51,4	104,9	89,5
program	2 stages	5,0	7,3	7,7	17,3	18,5	11,6	7,8	16,1	10,8	5,2	10,73
I.V.	digestion	84,5	93,3	101,2	107,1	125,4	99,3	97,4	95,5	53,1	98,4	95,5

e centrum
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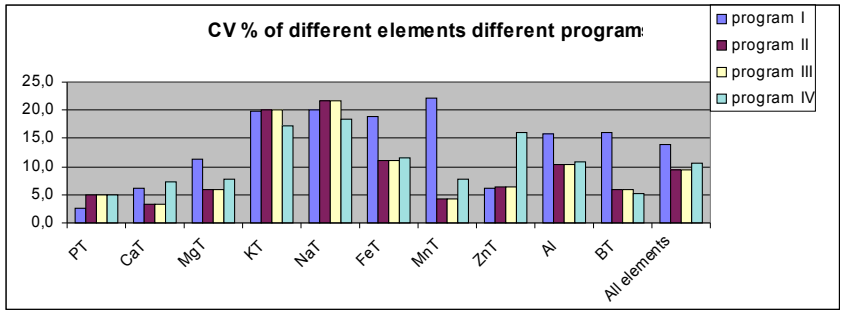
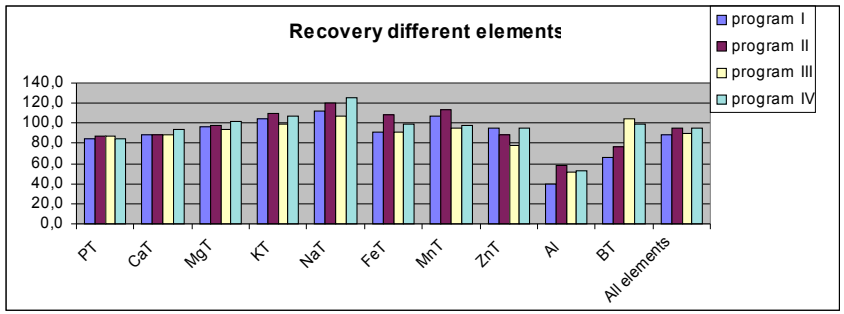
The comparison of CV % and Recovery of different methods





- We digested sample by four methods and determine all digested solution by ICP together in one round
- CV % of some elements was again too high e.g. K, Na over 20 %
- The recovery was too low for some sample e.g. P, Ca around 80 % and Al only 50 %

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Our results

- We evaluate the results using average value of CV% and recovery of all determined elements by relevant program
- The best average recovery was **95.5%** in Program IV (two stages digestion), but CV % was from 2.0 to 11 %
- Average variability of this digestion from all elements was **10.75 %**
- We did digestion different samples of foliage Wepal by this method

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Different samples

number	1.	2.	3.	4.	5.	6.	7.	8.	9.
sample	PT	CaT	MgT	KT	NaT	FeT	MnT	ZnT	AlT
	g/kg	g/kg	g/kg	g/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
124	2,34	22,7	1,63	26,1	199	224	40	19,9	271
V %	2,9	2,9	2,7	9,1	13,2	7,3	2,2	9,8	10,3
recovery	100	104	117	106	128	113	141	127	96
160	2,26	0,872	0,885	19,4	75	210	7,81	16	215
V %	1,3	1,2	2,2	1,5	4,0	0,9	2,8	5,0	15,6
recovery	102	118	121	96	134	131	271	136	89
949	2,03	11,4	1,6	23,3	242	205	20,1	103	192
V %	1,1	4,2	8,3	12,6	17,2	11,6	24,7	2,7	19,8
recovery	102	104	112	114	126	118	132	101	98
154	5,12	4,7	1,8	38,6	2790	325	79,8	39	144
V %	0,8	1,9	1,9	5,5	6,4	2,5	1,9	4,4	7,7
recovery	104	103	107	118	138	115	117	105	89

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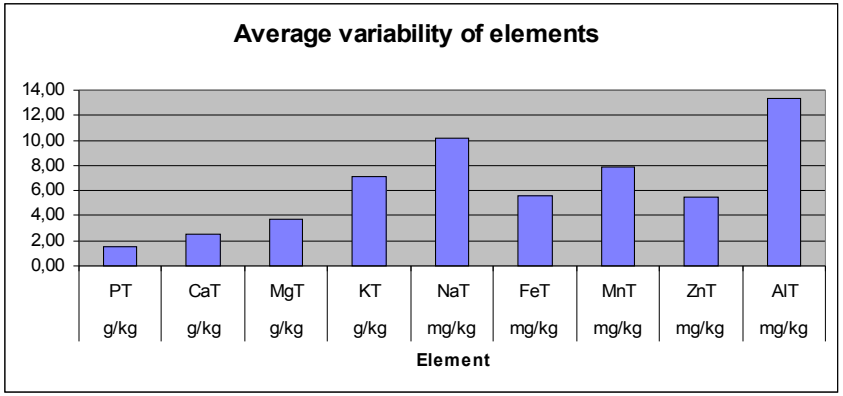
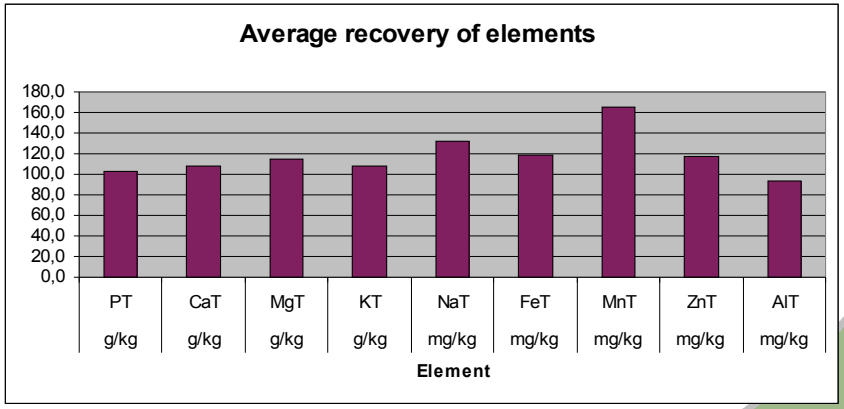
- **Chart 1** Average variability of the same element in different samples (only Na and Al had the higher CV than 10 %)
- **Chart 2** Average Recovery of the same element in different samples (elements Mg, Na, Fe, Mn and Zn had higher recovery than 110 %)
- The results were better but we are not satisfied
- So we hopeless search for next solution of our problem





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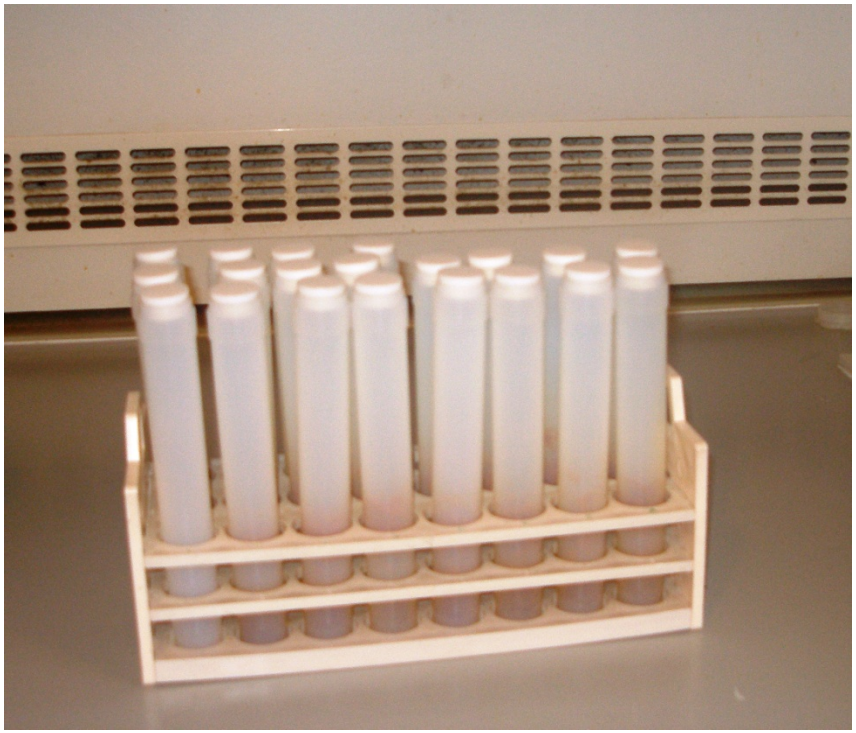

Cooperation with Comenius University

- Recommend us method for foliage
- Weight 0.500 g plant tissue (dry ground)
- Add 2 ml water let stand a few minutes
- Pour carefully 5 ml nitric acid observed intensity of foam
- Wait 10-20 minutes (when foaming finish) add 1.0 ml Hydrogen Peroxide (30 %)

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Cooperation with Comenius University

- Wait a few hours
- Better overnight leaving the vessel covered
- The next day do digestion
- Power 1200 W
- 100 % of power
- RAMP 20 minutes
- Temperature 150 C
- HOLD 10 minutes
- for 20 vessels cooling 30 minutes

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Cooperation with Comenius University

- Whole process is not so turbulent
- Necessary to observe carefully the intensity of foaming
- If it is too high add more hydrogen peroxide by drops
- Wait longer before digestion
- Increase Ramp of digestion about 5 (till 15 minutes)

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Evid. NO.	1.	2.	3.	4.	5.	6.	7.	8.	9.
sample	PT	CaT	MgT	KT	NaT	FeT	MnT	ZnT	CuT
	g/kg	g/kg	g/kg	g/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
ref.value	3,255	3,291	2,270	16,84	23,31	72,60	14,31	21,46	6,95
09-0394	3,083	3,376	2,206	14,526	27,491	72,09	15,386	22,057	6,78
%CV	3,66	5,48	0,96	2,62	13,41	25,18	4,88	34,38	12,59
recovery	94,7	102,6	97,2	86,3	117,9	99,3	107,5	102,8	97,6
ref.value	5,975	84,06	11,79	43,49	3076	360,3	218,7	79,60	10,10
09-0395	5,905	75,989	11,708	37,994	3035	332,4	226,4	72,62	9,72
%CV	5,12	6,23	1,47	5,07	4,20	3,69	1,41	20,98	5,25
recovery	98,8	90,4	99,3	87,4	98,7	92,3	103,5	91,2	96,2
ref.value	2,346	22,71	1,64	26,04	195	222,5	40,1	19,78	4,32
09-0396	2,302	23,07	1,64	22,863	195,5	218,6	41,97	22,841	4,68
%CV	3,49	4,52	1,12	2,62	6,76	11,83	1,85	35,09	12,17
recovery	98,1	101,6	99,9	87,8	100,3	98,2	104,7	115,5	108,5
ref.value	3,636	11,00	2,00	27,46	386	88,7	124,2	147,30	8,09
09-0397	3,36	10,71	2	24,089	365	87,5	130,2	136,2	8,46
%CV	7,82	7,39	1,57	3,07	2,88	5,78	2,99	15,69	
recovery	92,4	97,4	100,0	87,7	94,5	98,7	104,8	92,5	104,6

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Average CV% and Recovery Wepal 2/09

	PT	CaT	MgT	KT	NaT	FeT	MnT
Average	g/kg	g/kg	g/kg	g/kg	mg/kg	mg/kg	mg/kg
%CV	5,02	5,91	1,28	3,34	6,81	11,62	2,78
recovery	96,0	98,0	99,1	87,3	102,8	97,1	105,1

Our results Wepal 2/09

- We value the digestion of four samples Wepal and each sample digested six times parallelly
- CV improves for the most elements
- Recovery is the closer to 100 % except element K

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Results- Notes

- Digestion is only first step in process of determination elements
- Some problems could have been closely connected with old instrument ICP
- We check attentively cleanliness of digestion and preciseness of work
- Our results has been gradually improved

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Next problems ?

- It is difficult to find the optimal program for all sample of foliage
- We need a lot of patience and wisdom and some experience
- We still could not find answer for a lot of question
- But we hope that with your help to manage it

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Thanks for your attention

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