

MICROCALORIMETRIC SIMULATION STUDIES ON PHASE TRANSITIONS IN LIPIDS

Course of measurements:

1. Turn on the computer and click the icon „Biophys”.
2. Open the file „calorimeter” on the computer desk by clicking the left key of the mouse twice.

Part A. Studies on the phase transition of the lipid DMPC.

1. Press the white arrow in the upper left corner on the computer screen.
2. Press the key „configuration of measurements”. Then the configuration window will appear.
3. Select the following parameters for measurements:
 - a. window - „Name of the lipid” - select **DMPC**,
 - b. window - „Starting temperature” - set the value **285 K**,
 - c. window - „Final temperature” - set the value **315 K**.
 - d. window - „Cooling temperature” – set the value **280 K**.
 - e. window - „Scanning rate” – set the value **3 K/min**.
4. After having set the parameters press the key „Ready”

ATTENTION! Follow the instructions in the window „info” !

5. After having set the parameters:
 - a. An automatic cooling of the sample to the temperature set in the point 4d is starting. Cooling of the sample is to be done in order to eliminate all undefined states and disturbances before the actual measurement starts. When this temperature is reached press the key **”Start”**.
 - b. Heating of the sample with the scanning rate set in the point 4e is starting. When the “starting temperature” is reached the plot showing the enthalpy-to-temperature – $H(T)$ – relationship (thermogram) is starting to appear.
 - c. When the **“final temperature”** is reached an automatic cooling of the samples is taking place.
 - d. Press the key **„Analysis”**. Two cursors: the red one and the green one will then appear on the computer screen.

ATTENTION! When analyzing the results, please, perform individual actions (clicks) EXACTLY according to the instructions below. Any change may cause the program to hang, which will require you to repeat the scan from the beginning!

6. Using the computer mouse set:
 - a. The red cursor at the point where the last linear segment of the base line before the main phase transition peak starts. Then press the key **„BL before peak – start”**.
 - b. The red cursor at the point where the main phase transition peak starts. Then press the key **„BL before peak – end”**.
 - c. The green cursor at the point where the main phase transition peak ends. Then press the key **„BL after peak – start”**.
 - d. The green cursor at the point where the first linear segment of the base line after the main phase transition peak ends. Then press the key **„BL after peak – end”**.
 - e. Press the key **„Results”**
 - f. Then the parameters of the phase transition will appear on the computer screen. Write down these parameters to the work-sheet of the final report. Then press the key **„Exit”**.

7. Wait until the cooling of the sample is complete.

PART B. Studies on a phase transition of the lipid DMPG.

1. Press the key „configuration of measurements”.
2. Select the following parameters for measurements:
 - a. window - „Name of the lipid” - select **DMPG**,
 - b. window - „Starting temperature” - set the value **285 K**,
 - c. window - „Final temperature” - set the value **315 K**.
 - d. window - „Cooling temperature” – set the value **282 K**.
 - e. window - „Scanning rate” – set the value **3 K/min**.
 - f. After having set the parameters press the key „**Ready**”
3. Do the measurements such as in the part „A”.

PART C. Studies on a phase transition of the lipid DPPC.

1. Press the key „configuration of measurements”.
2. Select the following parameters for measurements:
 - a. window - „Name of the lipid” - select **DPPC**,
 - b. window - „Starting temperature” - set the value **300 K**,
 - c. window - „Final temperature” - set the value **330 K**.
 - d. window - „Cooling temperature” – set the value **290 K**.
 - e. window - „Scanning rate” – set the value **3 K/min**.
 - f. After having set the parameters press the key „**Ready**”
3. Do the measurements such as in the part „A”.

PART D. Identifying an unknown lipid.

1. Press the key „configuration of measurements”.
2. Select the following parameters for measurements:
 - a. window - „Name of the lipid” - select **“The unknown lipid”**,
 - b. window - „Starting temperature” - set the value **285 K**,
 - c. window - „Final temperature” - set the value **330 K**.
 - d. window - „Cooling temperature” – set the value **281 K**.
 - e. window - „Scanning rate” – set the value **4 K/min**.
 - f. After having set the parameters press the key „**Ready**”
3. Do the measurements such as in the part „A”.
4. Taking into account the previous results identify the unknown lipid.
5. Write down the name of this lipid to the work-sheet of the final report.
6. Plot the thermogram on a sheet of a plotting paper.

II. THEORETICAL PART

1. Redraw the exemplary thermogram $H(T)$ on the plotting paper. In the drawing mark pretransition and its temperature, main phospholipid phase transition and its temperature, half-height width of the peak, and transition enthalpy change.
2. Answer the questions from part II.2 of the report sheet.

Required theoretical knowledge:

1. Structure of biological membranes.
2. Phase transitions in lipids and their significance in biological systems.
3. Structural parameters of lipid molecules affecting their transition temperatures.
4. Calorimetry. Principles of work of a differential scanning microcalorimeter. Parameters of the thermogram (transition temperature, transition enthalpy change, half-width height).

Wrocław Medical University Department of Biophysics and Neuroscience	Practical No 9 Microcalorimetric simulation studies on phase transitions in lipids.	
..... Student names		Faculty: Group number: Date:
Grade:	Tutor's signature:	

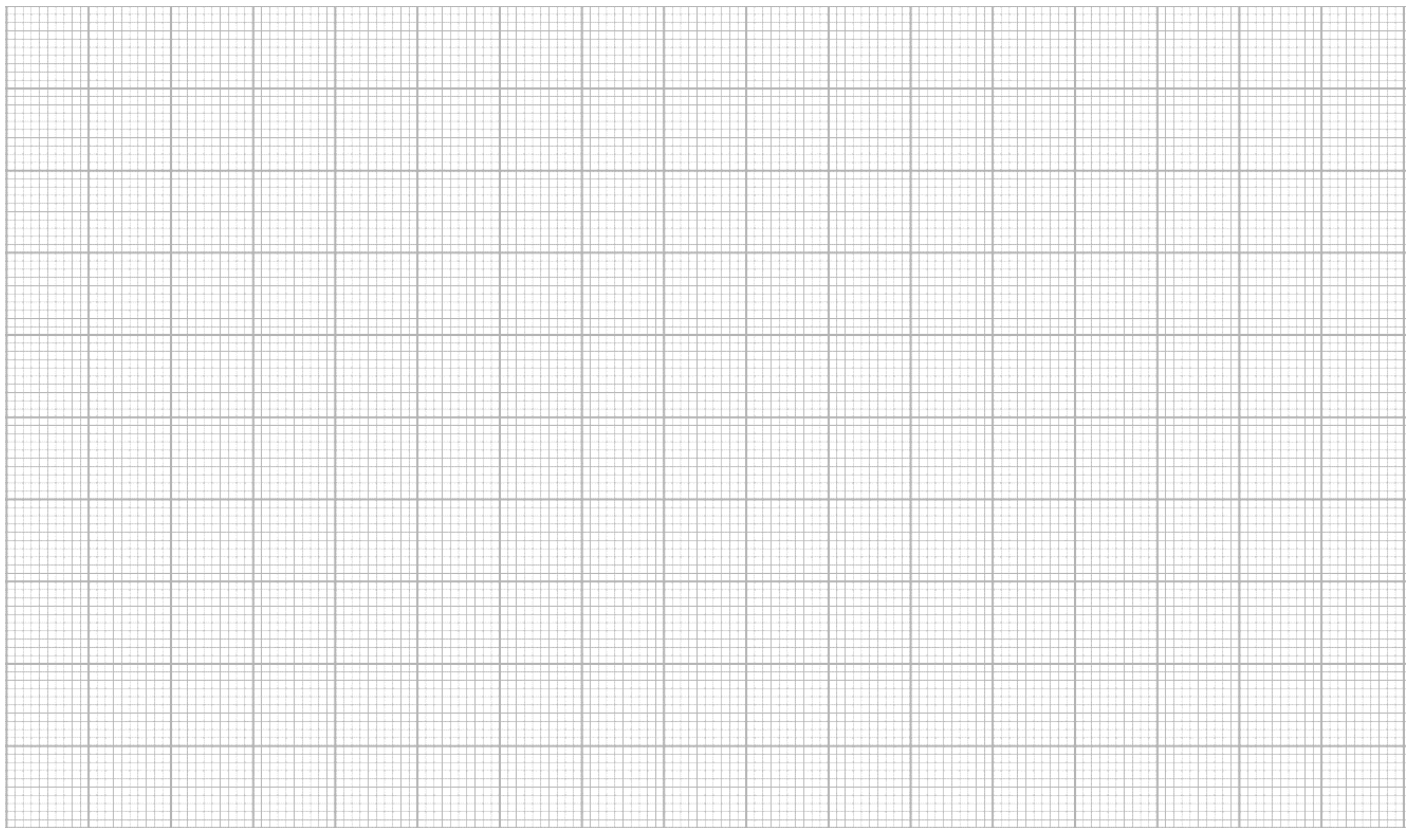
1. Perform the measurements according to the instructions.
2. Put the values of transition temperature, area under the transition peak and transition half-height width to the table below.

Sample No	Transition temperature [°C]	Area under the transition peak	Half-height width of peak [°C]
DMPC			
DMPG			
DPPC			
Unknown lipid			

Identificated lipid

II. THEORETICAL PART

1.A.



2.

A. What is phase transition?

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B. What phase transitions are typical for phospholipids?

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C. Describe the changes which lipid bilayer undergoes during pretransition?

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D. What does half-height width of the calorimetric peak inform us about?

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E. What is enthalpy?

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F. How can differential scanning calorimetry be used in pharmacology?

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