

Kinetics-like equation for cerebrospinal fluid-blood diffusion models

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ABSTRACT

Introduction: The diffusion of proteins from the blood to the cerebrospinal fluid is influenced by its molecular weight and by the intrinsic properties and biological properties of the protein.

Methods: Paired samples of serum and cerebrospinal fluid were taken from normal subjects to quantify albumin and proteins of the lectin pathway of the complement system. The distribution of these with regard to the value of $Q_{\text{Albumin}} = (\text{Albumin in serum} / \text{albumin in cerebrospinal fluid})$ was evaluated because this protein is used as a marker of the passage of the barrier.

Results: It was observed that some of these describe a saturation pattern which resembles the curves that describe the Michaelis-Menten reaction of enzymatic activity. This led to

the consideration of two constants that will help to characterize the behavior of these proteins by spreading to the cerebrospinal fluid: the maximum Q of the protein, which is the maximum proportion found empirically between the concentrations in blood and cerebrospinal fluid and the value Kcdw which is the value of the average diffusion speed of Q albumin when the semi-maximal value of the Q of the protein under study is obtained.

Conclusions: Empirically obtained constants will help the characterization and differentiation of the diffusion of these new proteins as they pass from the blood to the cerebrospinal fluid.

Keywords: cerebrospinal fluid-blood; Q albumin; proteins.

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INTRODUCTION

Complement system includes three pathways: classical, alternative and lectin pathway.

The last one is present since from the invertebrates, but it is not yet well defined. It is still discovering new structures and new evidences of its component's functions.

So far there are five different forms of pattern recognition (PRM): mannose binding lectin (MBL); H-, L- and M-ficolin (also known as Ficolin-3, -2 and -1, respectively); and collectin K (CL-K1): an MBL (MASP) -1 and -2 are associated with the serine protease to activate the complement. In addition, CL-K1 and collectin L 1 (CL-L1) are related to heteromers that are also associated with MASPs and activate complement. ⁽¹⁾

These initiators are not capable of initiating the enzymatic cascade by themselves; it needs other structures with the enzymatic characteristics that become the beginning of this activity, such as MASP2.

Although MASP-2 is capable of self-activation, recent studies have shown that MASP-1 is the exclusive activator of MASP-2 under physiological conditions. ^(1,2) MASP3 and the MAp44 are components with regulatory functions in this pathway.

These proteins are found in the highest percentage in the blood and they diffuse through the blood-brain barrier, but they can also be synthesized in the central nervous system.

These proteins participate in enzymatic reactions in the lectin pathway, where a balance is established between the activated and non-activated forms and between the soluble forms and those associated with other surfaces.

When the variation of Q albumin as a measure of protein diffusion between blood and CSF is plotted against the quotient diffusion of some of the lectin pathway, a typical curved curve is observed that resembles the enzymatic reactions shape, such as Michaelis-Menten reaction.

The objective of this work is to evaluate the behavior of the lectin pathway component MASP2, MASP3 and MAp44, based on their observed dynamics mimicking enzymatic activity.

METHODS

MASP2 was quantified in 56 serum and CSF samples by the enzyme-linked immunosorbent assay (ELISA) kits (Hycult Biotech, Uden, the Netherlands). ⁽³⁾

Plasma levels of MASP3 and Map44 were measured using time-resolved immunofluorometric in duplicates. All assays and specific antibodies for the assays were produced in-house. Detailed description for each assay can be found in the reference. ⁽⁴⁾

Albumin in serum and in CSF was quantified by nephelometric assay using Behring Nephelometer-Analyzer from Siemens (Marburg, Germany).

Statistical package MedCalc version 13.3.3.0 and GraphPad Prism version 5.01 was used to perform statistics and graphs.

RESULTS

The passage of proteins from the blood to the CSF is characterized from the values of Q Albumin. The distribution of the QMASP2, QMASP3 and QMap44 are shown in Fig. 1.

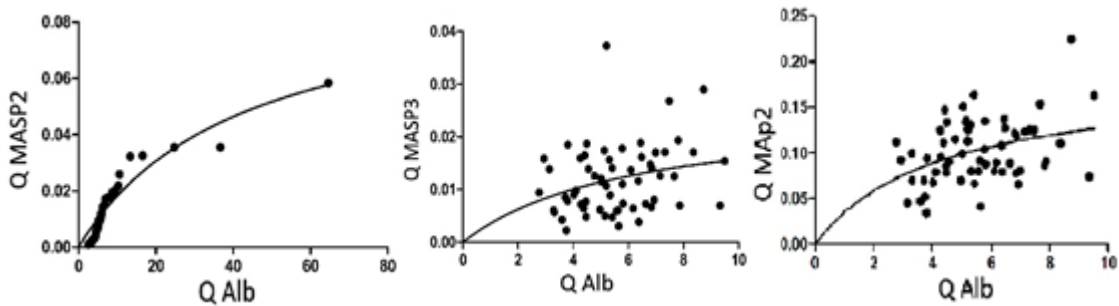


Fig. 1- Variation of QMASP2, MASP3 and QMap44 with Q albumin.

In the following table 1 you can see the new constants Q_{max} and K_{dacz} calculated from the similarity with the distribution of the Michaelis-Menten reaction.

Table 1- Kdacw and Q max of MSAP2. MASP3 and Map44

Contants	QMASP2	QMASP3	QMap44
Qmax	0,09569	0,02542	0,1836
Kdacw	42,64	6,125	4,294
Std. Error			
Qmax	0,01148	0,01339	0,04864
Kdacw	7,193	6,243	2,619
95 % Confidence Intervals			
Qmax	0,07265 to 0,1187	-0,001395 to 0,05223	0,08621 to 0,2810
Kdacw	28,21 to 57,07	0,0 to 18,63	0,0 to 9,538
Goodness of Fit			
Degrees of Freedom	54	58	58
R ²	0,8668	0,06656	0,1368
Absolute Sum of Squares	0,0008513	0,002362	0,06113
Sy.x	0,003970	0,006382	0,03247
Number of point analyzed	56	60	60

DISCUSSION

The passage of these components of lectin pathway from blood to CSF has allowed characterize these proteins as blood derived with the possibility of being synthesized in the CNS (1) because they follow the established statements for these types of proteins.

MASP2 has a behavior when it is distributed based on the speed of diffusion between the blood and the CSF that resembles a reaction of the Michaelis-menten type. This favors that this curve can be used to characterize this protein whose tracing resembles a reaction of enzymatic activity. The same thing happens with the regulation complement molecules MASP3 and MAp44.^(5,6)

By this way it was possible to characterize the new constants Kdacw and Qmax for each ones of these components of the lectin pathway.

The Q_{max} is the maximum value experimentally obtained from the diffusion of the protein when it passes from blood to CSF. It is the maximum relationship between these proteins when they pass from blood to CSF as a function of its serum concentration. It is the maximum diffusion speed between blood and CSF

The $K_{d_{acw}}$ is the Q value of the evaluated protein from the passage through the blood-LCR barrier that corresponds to the half-maximal speed-It can be experimentally obtained from $Q_{max} / 2$ and is a constant value depends on the molecule which is being evaluated.

With these new constants, the passage of these proteins from the serum to the CSF can be better defined and allows improving theoretical and practical analysis of this dynamics in blood-CSF barrier.

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