Serum complement activity in differentiating between bacterial and viral infections

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Introduction

The complement system is a humoral part of the immune system which consists of number of proteins that may help antibodies kill bacteria or lyse the pathogens on their own. It can be divided into classical-, lectin- and alternative-pathways all of which can contribute to the formation of the membrane-attack complex (MAC) which is responsible for the lysing of the cells. Microbe damage induces complement system activation.

Aim

The aim of this research was to see if the serum complement activity differs between serum samples with bacterial infections and with viral infections.
This was observed in response of the Luciferase modified *E. coli* (*E.coli-lux)* cells exposed to the diluted serum sample.

Materials and Methods

Research was conducted by measuring the bioluminescence of the *E. coli-lux* with high sensitivity luminescence assay at 37°C for 3 hours and 30 minutes after exposure to the serum.
Reaction was done in plate wells with 43350 bacteria and with serum levels of 0.5%; 0.25%; 0.125% and 0% as a control with a total volume of 100µl. HBSS was used as a buffer solution and E. coli cells were incubated in buffer solution for 20 minutes in RT before starting the measurement.
Complement activity was measured from frozen patient serums which were collected from TYKS ER and infectious diseases ward from suspected infectious patients with fever. Etiological diagnosis was conducted by doctor specialized in infectious diseases with microbiological, serological and radiological methods. 40 healthy volunteers were measured as a control.

Results

So far 166 serum samples with bacterial infection and 151 serum samples with viral infection have been measured. Bacterial infections had significantly more complement activity in comparison to viral infections and healthy controls.

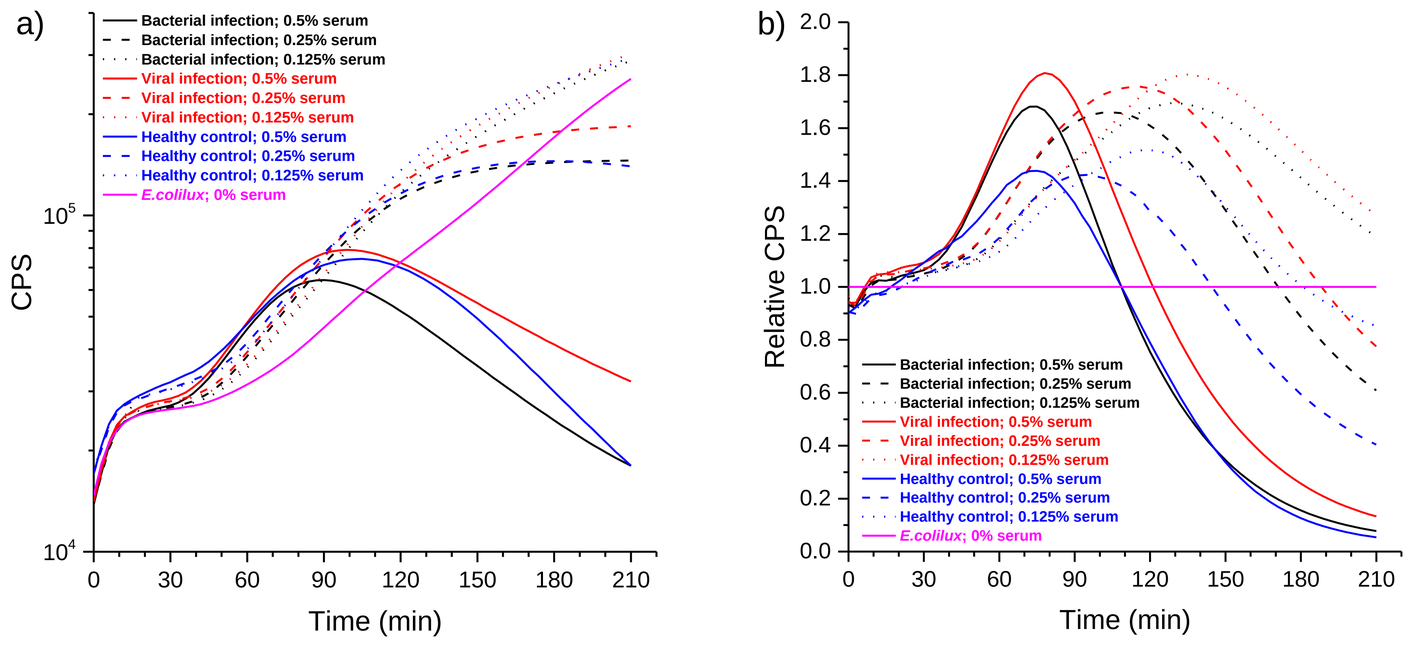

Figure 1. Bioluminescence levels (BL) of the 210 minutes measured. The black lines represent BL (counts per second, CPS) of the wells with the bacterial infected serum samples (BIS), the red lines the wells with the viral infected serum samples (BIV) and blue lines the healthy controls. Violet line represents baseline BL of the *E. coli-lux* in the buffer solution. a) Represents BL levels as a function of time. b) Represents the BL levels of the different dilutions in relation to the baseline calculated by BL (dilution)/BL (0% serum) as a function of time. As a result we can see that BIS peaks earlier in activity than BIV.

Table 1. Peak times and relative bioluminesence levels (rCPS) starting at 60 min mark and taken every 30 min of both bacterial and viral infections. Bacterial infections have significant differences to viral infection in 0.5% serum samples. An unpaired t-test was used to compare the means of two unrelated groups of samples.

0.5% serum sample					
	BACTERIAL INFECTION		VIRAL INFECTION		
	mean	SD	mean	SD	T-test
(peak-time - 40) (min)	52	23	62	24	p < 0.001
relative (r) CPS (60 min)	1.53	0.26	1.56	0.26	nsd
relative (r) CPS (90 min)	1.46	0.50	1.70	0.44	p < 0.001
relative (r) CPS (120 min)	0.75	0.53	1.02	0.52	p < 0.001
relative (r) CPS (150 min)	0.35	0.39	0.52	0.43	p < 0.001
relative (r) CPS (180 min)	0.16	0.25	0.26	0.29	p = 0.001
relative (r) CPS (210 min)	0.08	0.17	0.13	0.19	p = 0.008

Conclusions

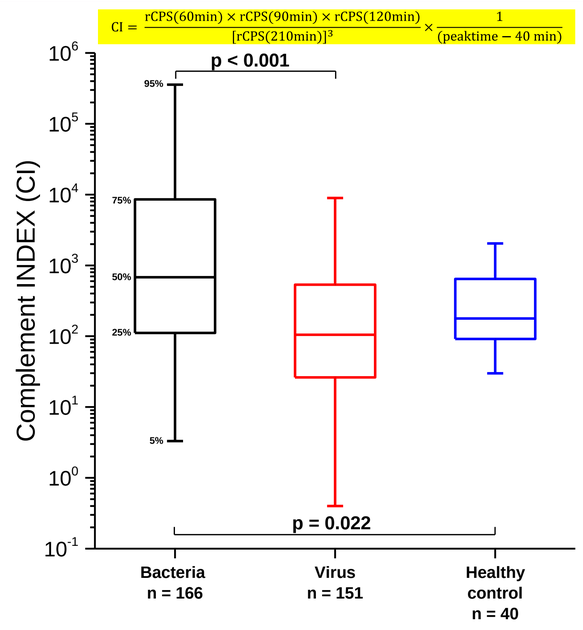

Figure 2. Complement INDEX (CI) of individual bacterial, viral and healthy patient samples represented as boxplot. CI (yellow box) formula represents significant differences between bacterial and viral samples. The Bonferroni corrected p-values of post-hoc pairwise between group comparisons were determined after the Kruskal-Wallis test.

Although the difference is significant, the method cannot be used on its own as a diagnostic tool to differentiate the infections as there is too much overlap.

References

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