

1 **Salocor SARS-CoV-2 Antigen Quantitative Assay Kit<sup>®</sup> (Salofa Ltd, Salo, Finland)**

2 **protocol**

3 The evaluated Salocor SARS-CoV-2 Antigen Quantitative Assay Kit<sup>®</sup> is based on a double  
4 antibody sandwich method. The kit contains a 96-well plate coated with anti-SARS-CoV-2 N  
5 protein antibody. If the serum sample contains SARS-CoV-2 N protein, the N protein  
6 attaches both solid phase anti-SARS-CoV-2 N protein antibody and biotin-labelled anti-  
7 SARS-CoV-2 N protein antibody added with serum sample. The test procedure is briefly as  
8 follows: 50 ul of buffer solution containing biotin-labelled antibody against SARS-CoV-2 N  
9 protein was added in each well coated with antibody against SARS-CoV-2 N protein. Next  
10 50 ul of serum sample was added to each well with the exclusion of control well and 5 wells  
11 for calibrators included in the kit and added alongside the sample material. After brief  
12 mixing, the plate was incubated at 37 °C for 60 minutes followed by washing the wells five  
13 times with the washing buffer. 100 ul of horseradish peroxidase-labelled streptavidin was  
14 added to each well. After brief mixing, the plate was incubated at 37 °C for 30 minutes  
15 followed by the washing step and adding 50 ul of buffer solution containing peroxidase and  
16 50 ul of buffer solution containing 3,3',5,5'-tetramethylbenzidine. After brief mixing, the  
17 plate was incubated at 37 °C for 15 minutes and color reaction was stopped by adding 50 ul  
18 of stop solution containing sulfuric acid. Absorbance values were measured at 450 nm with a  
19 reference set at 630 nm. The concentration values of samples were then calculated using  
20 standard curve with binomial fitting based on different calibrator absorbance values with  
21 known SARS-CoV-2 N protein concentrations (pg/ml). Concentration values  $\geq 2,97$  pg/ml  
22 were interpreted as positive according to the manufacturers' instructions.