

# DIN EN 14476:2025-11 (E)

## Chemical disinfectants and antiseptics - Quantitative suspension test for the evaluation of virucidal activity in the medical area - Test method and requirements (Phase 2/Step 1)

---

<b>Contents</b>	<b>Page</b>
European foreword .....	4
Introduction .....	6
1 Scope.....	7
2 Normative references.....	7
3 Terms and definitions.....	8
4 Requirements.....	9
5 Test methods .....	10
5.1 Principle.....	10
5.2 Materials and reagents, including cell cultures.....	11
5.2.1 Test viruses.....	11
5.2.2 Culture media, reagents and cell cultures.....	12
5.3 Apparatus and glassware.....	16
5.4 Preparation of test organism suspensions and product test solutions.....	17
5.4.1 Test organisms suspensions (test virus suspension).....	17
5.4.2 Product test solutions.....	18
5.5 Procedure for assessing the virucidal activity of the product.....	18
5.5.1 General.....	18
5.5.2 Test procedure.....	19
5.5.3 Modified method for ready-to-use products.....	21
5.5.4 Cytotoxicity caused by product test solutions.....	21
5.5.5 Interference control – control of cell susceptibility.....	22
5.5.6 Control of efficiency of suppression of product's activity.....	23
5.5.7 Reference test for virus inactivation.....	23
5.5.8 Titration of the virus control.....	25
5.5.9 Titration of test samples.....	25
5.6 Experimental data and calculation.....	25
5.6.1 Protocol of results.....	25
5.6.2 Calculation of infectivity titre (TCID <sub>50</sub> or PFU) and reduction.....	25
5.7 Verification of the methodology and test validity.....	33
5.8 Expression of results.....	34
5.8.1 General.....	34
5.8.2 Calculation of the virucidal activity of products.....	34
5.8.3 Control of active and non-active product test solution.....	34
5.9 Test report.....	35
Annex A (informative) Examples of viruses sorted according to their presence in the human body in case of virus infection.....	37
Annex B (informative) Detoxification of test mixtures by molecular sieving.....	39
B.1 Molecular sieving with Sephadex <sup>TM</sup> LH 20.....	39
B.1.1 Principle.....	39
B.1.2 Sephadex suspension.....	39
B.1.3 Procedure.....	39

<b>B.2</b>	<b>Molecular sieving using MicroSpin™ S 400 HR.....</b>	<b>41</b>
<b>Annex C</b>	<b>(informative) Example of presentation of data results for one active concentration .....</b>	<b>42</b>
<b>Annex D</b>	<b>(informative) Quantitative determination of formaldehyde and PAA concentrations ....</b>	<b>45</b>
<b>D.1</b>	<b>Quantitative determination of formaldehyde .....</b>	<b>45</b>
<b>D.2</b>	<b>Determination of PAA concentration .....</b>	<b>46</b>
<b>Annex E</b>	<b>(informative) Preparation of the glutaraldehyde test solutions (v/v).....</b>	<b>49</b>
<b>Bibliography</b>	<b>.....</b>	<b>50</b>