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Evaluation of official instrumental methods for the determination of particulate matter contamination in large volume parenteral solutions

L. MONTANARI*, F. PAVANETTO, B. CONTI, R. PONCI AND M. GRASSI†

*Department of Pharmaceutical Chemistry and †Institute of Applied Health Sciences, University of Pavia, Pavia, Italy

The distribution pattern of particle contamination in nine different types of LV parenteral solutions and the possibility of correlating the counts made with two official instruments (Coulter Counter and HIAC) were studied. Two hundred containers of LV parenteral solutions (corresponding to 40 batches) produced in Italy, were sampled. Each bottle was submitted to HIAC and Coulter Counter countings, for particle sizes ranging between 2 and 25 μm. For about 50% of the products, the two straight lines that represent the distribution of particle contamination obtained with the two methods did not cross-over within the studied size range, the Coulter Counter counts always proving higher than the HIAC ones. In the other cases, the cross-over point of the two lines occurred at varying size levels. Statistical analysis of the results pointed to a relationship between the contamination values obtained with the two counting methods for sizes ranging between 2 and 5 μm, but there was no correlation for sizes equal to, or higher than, 10 μm. From the maximum contamination levels established by the BP and the FU IX for the HIAC method, the corresponding values were calculated for the Coulter Counter method. Similarly the values were calculated the HIAC method based on the maximum values set for the Coulter Counter.

It is generally accepted that the extent of contamination of large volume parenterals by particulate matter requires to be controlled. The determination of the number and size of particles in liquids may be made microscopically or by using automatic electronic particle counters, which, because of their advantages, are preferred for routine quality control purposes. Official requirements relating to particulate matter in parenteral products are now addressed to the use of the Coulter Counter and HIAC instruments.

It is well known that the contamination values obtained with the available techniques for counting particles in solution may differ, because of the different particle parameters measured by the instruments. For this reason the British Pharmacopoeia (1980) has set the limit values of particulate matter according to the type of instrument used. For the Coulter Counter method the contamination limit values are: $\geqslant$1000 particles $ml^{-1} > 2$ μm and $\geqslant$100 particles $ml^{-1} > 5$ μm, whereas for the HIAC method they are: $\geqslant$500 particles $ml^{-1} > 2$ μm and $\geqslant$80 particles $ml^{-1} > 5$ μm.

By applying these evaluation criteria we have encountered wide differences in the quality evaluation of products. The Italian Pharmacopoeia requirement (1985) proposes the use of a light blockage instrument for particle count: other methods are permitted only provided that contamination limit values are correlated with the official ones. For the HIAC method the limit values are: $\geqslant$100 particles $ml^{-1} \geqslant 5$ μm, and $\geqslant$4 particles $ml^{-1} \geqslant 20$ μm.

Since different methods may be used to evaluate particulate matter, there is a need for correlation criteria. Many studies have been devoted to this subject. Groves & Wana (1977) analysed the same solutions (0·9% w/v NaCl) by the Coulter Counter and the light blockage methods. The results they obtained, when plotted on a log/log size distribution basis, crossed over at a size threshold of around 6 μm. Theoretically, a relation between the counts obtained with the Coulter Counter and with the light blockage instrument was calculated, taking into account the influence of the shape factor.

Haines-Nutt (1983) and Haines-Nutt & Munton (1984) confirmed these data for salt solutions, but not for solutions containing sugar or sugar derivatives. Other authors (Dawes et al 1983; Taylor & Spence 1983) found a low correlation and a wide variation among the results obtained by these two instrumental methods.

These studies were limited in extent by the number of solutions and samples examined. It therefore

* Correspondence.

seemed worthwhile to make a systematic investigation of the most common examples of LV parenteral solutions in numbers of units to give reliability to the results of statistical analysis. In total we examined 185 bottles of LV parenteral solutions from 37 different batches and from 8 pharmaceutical firms, and 15 bottles of normal saline prepared in our laboratory; Table 1 lists the products. The level of particulate contamination was determined by sampling each container with both Coulter Counter and HIAC method.

The statistical analysis of the results enabled us to study the possible correlation among the values obtained with the two methods. Also, the particle distribution-particle size relationships and acceptability criteria of the British and the Italian Pharmacopoeias were verified and discussed. This investigation was made with the aim of defining the contamination limits, as a function of the instrument used in such a way as to ensure the reproducible quality of a given LV product.

METHODS

Material and instruments for the determination of particulate matter in parenteral solutions

The instruments used were: HIAC/ROYCO model 3000, fitted with an HR60H sensor, with standard size range of 1–60 μm flow rate 10 ml min^{-1}, and supplied by the manufacturer already calibrated with standard spherical materials, and a Coulter Counter, model TAII, fitted with a 70 μm orifice tube. Calibration was with 8·7 μm diam. latex suspensions added to solutions previously filtered through 0·45 and 0·22 μm pore filter membranes. These solutions were used as such or diluted with 1·8% w/v sodium chloride solution.

The samples examined were from 185 containers of large volume (LV) solutions for parenteral use, from 37 batches and produced by eight commercial sources, and also from 15 bottles of 0·9% w/v sodium chloride solution, prepared in our laboratory, divided into 3 groups of 5 bottles each representing batches no. 38, 39, 40 (producer i, see Table 1). Also prepared were 0·9 and 1·8% NaCl solutions, filtered through Millipore membranes (pore size 0·45 and 0·22 μm). These were used for diluting non-conductive or viscous solutions, for analysis with the Coulter Counter. These solutions, when examined with the HIAC device, had not more than 10 particles ml^{-1} with diameter over 5 μm, and no particulate matter over 20 μm. When examined with the Coulter Counter, the particulate contamination was not more than 50 particles ml^{-1} with a diameter of more than 2 μm, and not more than 20 particles ml^{-1} with a diameter of more than 5 μm.

Suspensions of monodimensional polystyrene microspheres were used of the following average sizes: 2·87, 5·2, 8·7, and 19·1 μm (Coulter Electronics, Industrial Division, Hialeah, Florida).

Preparation of standardized latex suspension

The suspensions of the standard material used to verify calibration were prepared by diluting a few drops of a given latex suspension, supplied by the Coulter Electronics Co., with an adequare volume of salt solution filtered as above. The microsphere latex suspensions obtained were vigorously stirred and ultrasonicated for 1 min to eliminate the air bubbles present.

Samples preparation and analysis

Five bottles from each batch were examined. Table 1 reports the mean values (with s.d.) of the contamination values found, as the cumulative number of particles ml^{-1}.

Solutions were mixed by cautiously turning the bottles upside down twice. After removing the metal support, the surface of the container was washed by spraying water filtered through a 0·45 μm membrane

The contents of each bottle were distributed between the Coulter Counter container and the HIAC container.

For the 250 ml bottles, analysis with the HIAC counter was made directly in the original containers.

To eliminate air bubbles, samples to be examined were ultrasonicated for 1 min and allowed to stand for at least 10 min before being counted. During the reading of data, the solution under examination was stirred at a very low speed.

Analysis using the Coulter Counter method

Sample analysis and the determination of contamination values for each solution were made according to the model of Montanari et al (1982). For each bottle, 3 × 2 ml samples were examined.

Analysis using the HIAC device

Countings were made at different size levels (2, 3·5, 5, 10, 20, and 25 μm).

Analysis of whole bottles of the different solutions showed that the distribution of particulate matter was uniform in each bottle, at these contamination

Table 1. Cumulative particle number ml^{-1} (mean of 5 bottles per batch and standard deviation).

Batch No	Mfg	Solution	≥ 2 μm		≥ 3·5 μm		≥ 5 μm		≥ 10 μm		≥ 20 μm		≥ 25 μm	
			Coulter Counter	HIAC	Coulter Counter	HIAC	Coulter Counter	HIAC	Coulter Counter	HIAC	Coulter Counter	HIAC	Coulter Counter	HIAC
1	a	NaCl 0·9%	1349	209	316	81	77	37	8	6	0·4	0·4	0·2	0·2
			(404)	(109)	(131)	(41)	(33)	(18)	(3)	(4)	(0·1)	(0·1)	(0·2)	(0)
2	a	NaCl 0·9%	4750	490	1332	111	261	36	14	3	0·4	0·1	0·1	0·1
			(466)	(115)	(15)	(7)	(65)	(4)	(4)	(1)	(0)	(0)	(0)	(0)
3	b	NaCl 0·9%	566	44	64	14	9	6	1	1	0·1	0	0	0
			(748)	(18)	(57)	(5)	(6)	(2)	(0)	(1)	(0·1)	(0)	(0)	(0)
4	c	NaCl 0·9%	650	109	133	28	24	11	3	2	0·3	0·1	0	0
			(137)	(25)	(37)	(3)	(9)	(1)	(2)	(1)	(0·5)	(0)	(0)	(0)
5	g	NaCl 0·9%	152	75	50	27	13	10	1	1	0	0	0	0
			(10)	(2)	(1)	(1)	(3)	(1)	(1)	(0·1)	(0)	(0)	(0)	(0)
6	d	Replacement electrolyte solution	184	78	47	38	13	20	3	6	0·2	0·5	0·2	0·2
			(62)	(37)	(22)	(18)	(8)	(13)	(1)	(5)	(0·1)	(0·3)	(0·7)	(0·2)
7	d	Replacement electrolyte solution	2686	158	269	58	56	26	4	3	1	0·2	0·2	0·1
			(2541)	(78)	(133)	(29)	(32)	(11)	(2)	(2)	(0·2)	(0·1)	(0)	(0)
8	f	Replacement electrolyte solution	394	83	131	37	37	18	5	4	0·3	0·5	0·1	0·2
			(201)	(62)	(57)	(30)	(16)	(16)	(2)	(4)	(0·3)	(1)	(0·1)	(0·4)
9	f	Replacement electrolyte solution	534	78	177	32	51	13	7	2	0·5	0·2	0·2	0·1
			(303)	(57)	(83)	(24)	(22)	(8)	(4)	(1)	(0·4)	(0·1)	(0·2)	(0·1)
10	g	Ringer lactate	256	63	91	28	32	14	6	3	0·5	0·4	0·1	0·2
			(70)	(29)	(25)	(11)	(12)	(5)	(3)	(1)	(0·4)	(0·1)	(0·1)	(0·1)
11	b	Ringer lactate	898	130	220	50	58	26	8	5	2	0·3	2	0·1
			(103)	(27)	(39)	(18)	(16)	(13)	(4)	(3)	(3)	(0·3)	(2)	(0)
12	b	Amino acids 3·5%	1317	186	493	78	145	35	12	6	1	0·4	0·1	0·1
			(439)	(34)	(138)	(12)	(38)	(5)	(6)	(2)	(0)	(0·1)	(0)	(0)
13	d	Amino acids 5%	533	111	180	45	51	21	3	5	0	0·5	0	0·1
			(173)	(77)	(64)	(33)	(16)	(16)	(1)	(5)	(0)	(1)	(0)	(0·2)
14	g	Amino acids 8·5%	331	88	107	34	25	16	2	2	1	0·2	0·1	0
			(81)	(10)	(29)	(2)	(8)	(1)	(2)	(1)	(0)	(0)	(0)	(0)
15	h	Liophilized amino acids in NaCl 0·9%	1118	273	322	128	75	63	5	11	0	1	0	0·6
			(156)	(82)	(72)	(52)	(25)	(28)	(3)	(5)	(0)	(1)	(0)	(0·3)
16	a	Dextrose 33%	4842	149	893	51	151	19	16	2	2	0·1	1	0
			(2545)	(75)	(324)	(25)	(31)	(10)	(2)	(1)	(0·4)	(0)	(0·2)	(0)
17	b	Dextrose 33%	322	12	62	4	14	2	8	0·4	1	0	0·4	0
			(235)	(0·3)	(78)	(1)	(20)	(0·4)	(11)	(0)	(1)	(0)	(0)	(0)
18	g	Dextrose 10%	670	229	218	24	52	8	4	2	0·3	0·3	0	0·2
			(160)	(249)	(51)	(14)	(12)	(5)	(2)	(2)	(0·3)	(0·2)	(0)	(0·1)
19	b	Dextrose 10%	713	110	177	33	33	15	5	6	1	1	0·3	0·2
			(612)	(110)	(160)	(38)	(26)	(17)	(2)	(4)	(1)	(0·2)	(0·4)	(0·2)
20	g	Dextrose 5%	394	47	134	19	35	9	5	2	1	0·1	0	0·1
			(66)	(18)	(25)	(7)	(8)	(3)	(2)	(1)	(1)	(0)	(0)	(0)
21	d	Dextrose 5%	287	113	73	36	19	16	1	4	0·2	0·1	0	0
			(97)	(26)	(30)	(7)	(10)	(4)	(0·5)	(1)	(0·2)	(0)	(0)	(0)
22	b	Dextrose 5%	482	36	146	12	42	5	9	1	0	0	0	0
			(271)	(18)	(139)	(8)	(42)	(4)	(9)	(1)	(0)	(0)	(0)	(0)
23	g	Dextrose 5% in NaCl 0·9%	361	44	82	18	18	9	3	3	0·2	0·2	0	0
			(139)	(23)	(42)	(11)	(13)	(6)	(3)	(2)	(0·3)	(0·2)	(0)	(0·1)
24	c	Mannitol 18%	1206	378	304	38	60	17	10	3	0·2	0	0·1	0
			(198)	(632)	(58)	(25)	(18)	(12)	(6)	(3)	(0·3)	(0)	(0·2)	(0)
25	b	Mannitol 18%	488	178	57	96	14	59	7	17	2	4	0·5	2
			(113)	(106)	(17)	(72)	(11)	(49)	(5)	(17)	(1)	(4)	(1)	(3)
26	a	Mannitol 18%	601	615	177	314	38	174	4	32	0·2	4	0·2	2
			(401)	(425)	(144)	(244)	(34)	(144)	(5)	(29)	(0·2)	(3)	(0·3)	(1)
27	d	Mannitol 10%	461	83	137	29	37	11	8	2	0·4	0·1	0·1	0
			(190)	(63)	(54)	(27)	(16)	(9)	(6)	(1)	(1)	(0)	(0·1)	(0)
28	b	Mannitol 10%	325	65	93	24	25	10	7	2	1	0·1	0·1	0
			(73)	(46)	(23)	(15)	(5)	(5)	(1)	(1)	(0·3)	(0·1)	(0·2)	(0)
29	g	Mannitol 10%	269	82	50	30	12	13	3	2	0·3	0·1	0·2	0·1
			(212)	(21)	(73)	(6)	(20)	(1)	(5)	(0·3)	(0·5)	(0·1)	(0·3)	(0·1)
30	g	Fructose 10%	197	54	57	21	13	9	3	2	0·3	0·1	0·2	0
			(103)	(18)	(33)	(4)	(9)	(1)	(3)	(0·4)	(0)	(0·1)	(0)	(0)
31	b	Fructose 10%	85	40	29	19	14	10	5	3	0·5	0·2	0·3	0·1
			(35)	(32)	(16)	(12)	(12)	(5)	(4)	(1)	(0·2)	(0·1)	(0·2)	(0)
32	d	Fructose 10%	1001	352	264	83	37	28	3	5	1	0·1	0·4	0
			(382)	(306)	(52)	(93)	(7)	(39)	(2)	(9)	(1)	(0·2)	(0·4)	(0)
33	g	Fructose 5%	354	93	111	35	26	17	5	4	0·2	0·1	0	0
			(53)	(43)	(19)	(16)	(6)	(7)	(0·3)	(4)	(0)	(1)	(0)	(0)
34	e	Dextran 40 in NaCl 0·9%	666	121	164	43	46	21	8	5	1	1	0·3	0·2
			(253)	(45)	(54)	(8)	(12)	(3)	(3)	(1)	(1)	(0·2)	(0·2)	(0·1)
35	g	Dextran 40 in NaCl 0·9%	201	30	62	12	17	6	2	2	0·5	0·2	0·1	0
			(22)	(4)	(6)	(1)	(4)	(1)	(1)	(0·3)	(1)	(0·2)	(0)	(0)
36	g	Dextran 70 in NaCl 0·9%	210	20	61	10	16	6	3	2	1	0·2	0·5	0·1
			(48)	(2)	(10)	(1)	(5)	(1)	(4)	(0·3)	(1)	(0·1)	(1)	(0·1)
37	c	$NaHCO_3$ 1·4%	7104	1814	2469	664	610	251	29	13	0·3	0·2	0	0·1
			(1574)	(304)	(396)	(263)	(140)	(131)	(14)	(7)	(0·2)	(0·1)	(0)	(0)
38	i	NaCl 0·9%	216	80	62	33	18	17	5	5	0·5	0·5	0·1	0·3
			(82)	(29)	(22)	(13)	(6)	(8)	(2)	(2)	(0·2)	(0·3)	(0·1)	(0·2)
39	i	NaCl 0·9%	168	63	58	28	28	14	4	4	0·2	0·4	0	0·2
			(95)	(43)	(31)	(22)	(24)	(11)	(2)	(3)	(0·1)	(0·3)	(0)	(0·2)
40	i	NaCl 0·9%	230	132	58	57	16	29	3	7	0·3	0·5	0·1	0·2
			(116)	(96)	(24)	(43)	(6)	(22)	(2)	(5)	(0·2)	(0·3)	(0·1)	(0·2)

levels (Pavanetto et al 1986). Therefore, only 3 × 10 ml samples of each solution were examined.

RESULTS AND DISCUSSION

Cumulative number of particles–particle size distribution and coincidence point of the values obtained with the HIAC and Coulter Counter methods

In all the solutions examined the cumulative number of particles–particle size relation obtained with both methods follows a log–log distribution, according to examples reported in the literature (Groves 1973; Groves & Wana 1977; Taylor & Spence 1983; Montanari et al 1982; Haines-Nutt & Munton 1984).

The correlation coefficients found for 6 pairs of log cumulative number–log size values (2, 3·5, 5, 10, 20, 25 μm) ranged between 0·95 and 0·99.

Through linear regression analysis of the values obtained, for each solution the equation of two straight lines and their cross-over point was calculated.

Groves & Wana (1977) reported that in the case of saline solution the distribution of contamination obtained by the Coulter Counter and HIAC methods, when plotted on a log–log size distribution basis, crossed over at a size threshold of around 6·0 μm (4·2–6·8 μm). Usually, at sizes below 6·0 μm the values yielded by the Coulter Counter were higher than those yielded by the HIAC method, while at sizes exceeding 6·0 μm the opposite occurred.

Moreover, a correlation among the two techniques was shown for ionic solutions, but not for sugar-containing solutions (Haines-Nutt 1983; Haines-Nutt & Munton 1984).

The British Pharmacopoeia has set a maximum number for particulate matter depending on the instrument used, thereby establishing a relationship between the numbers for particulate matter obtained with the Coulter Counter and those obtained with the HIAC device for all the types of solution subject to control.

An analysis of results showed the Coulter Counter and HIAC distributions to cross over in the 4·2–6·8 μm range only for solution no. 6. For about 50% of the products, HIAC values were lower than Coulter Counter values, at all the examined sizes. In the other cases, the crossover point was found to be at a size level below 4·2 μm (10%) or between 6·8 and 25 μm (37%). In one exceptional case (batch no. 26) the HIAC counts were always higher than the Coulter Counter values.

Furthermore, the ln CC–ln HIAC linear relationships at different size levels showed no statistically significant difference depending on product nature, according to our data.

Statistical analysis

To assess the correlations among the HIAC and Coulter Counter counting techniques, the following analyses were performed, in the order: preliminary univariate analysis, simple-multiple regression analysis (see Draper & Smith 1981), regression diagnostic and unit analysis (Weinsberg 1980).

The units considered in the analysis reflect the mode of data collection and concern the 40 batch-units, obtained from the calculation of the values (means with s.d.) of 5 bottles per batch, containing different solutions and from different manufacturers. For each batch, the mean values and the s.d. relative to the different particle sizes (2, 3·5, 5, 10, 20, 25 μm), as evaluated with the two methods, are reported in Table 1, together with the type of solution and indication of manufacturer.

To allow the correct use of the statistical method chosen (which assumes the linearity of the variables), the original values have been re-expressed logarithmically, the relationships being assumed to be linear for the logarithm values and multiplicative for the original ones. The preliminary analysis showed the statistical irrelevance of the ln HIAC–ln CC relationship for diameters exceeding 10 μm.

The correlation coefficients are, respectively, $r_{d \geq 3.5} = 0.66$, $r_{d \geq 2} = 0.59$, $r_{d \geq 5} = 0.22$.

The ln HIAC–ln CC relationship for the 2, 3·5 and 5 μm particle sizes is shown in Table 2.

The procedure used for the determination of the ln CC = f (ln HIAC) + e, ln HIAC = f (ln CC) + e equations at the different particle sizes D ≥ 2, ≥ 3·5,

Table 2. ln HIAC and ln CC relationships and corresponding statistics for the 2, 3·5 and 5 μm threshold sizes.

D ≥ 2 μm	
ln (CC) = 2·62 + 0·74 ln (HIAC)	ln (HIAC) = 0·39 + 0·66 ln (CC)
R^2 = 0·62 n = 40	R^2 = 0·721 n=37
S = 0·669 vs Sy = 1·067	S = 0·757 vs Sy = 1·413
D ≥ 3·5 μm	
ln (CC) = 2·32 + 0·69 ln (HIAC)	ln (HIAC) = 1·323 + 0·456 ln (CC)
R^2 = 0·74 n = 40	R^2 = 0·947 n = 37
S=1 vs Sy = 1·924	S = 0·931 vs Sy = 3·982
D ≥ 5 μm	
ln (CC) = 1·89 + 0·50 ln (HIAC)	ln (HIAC) = 0·975 + 0·481 ln (CC)
R^2 = 0·42 n=40	R^2 = 0·876 n = 37
S = 1·268 vs Sy = 1·638	S = 1·251 vs Sy = 3·498

D : threshold size.
R^2 : the square of the correlation coefficient, which provides a measure of accuracy of prediction or the association between the response and predictor variables.
n : batch units.
S : estimated residual standard deviation of response variable.
Sy : standard deviation of response variable.

≥ 5 μm was as follows: (i) in a preliminary analysis the 'dummy' (Weinsberg 1980) terms relative to the 'type of solution' variable were considered, in view of the importance attributed in the literature to such a variable. The effect of that variable, however, bears no statistical relevance to our data and as a consequence it is not taken into account in the final equation. (ii) In view of the nature of the batch-units (mean of 5 bottle units), mean units were weighted for the reciprocal of the corresponding standard deviations of the response variable, so as to balance the influence of the most widely variable units (Weinsberg 1980). The weights are ln (s.d. CC)$^{-1}$ if the equation is ln CC = f (ln HIAC) + e and ln (s.d. HIAC)$^{-1}$ if the equation is ln (HIAC) = f (ln CC) + e. (iii) Some batch-units, if particularly influential at their extreme values, were omitted in the final equation (Cook 1979). Batches nos 5, 17, 31 were not included in the calculation of the ln HIAC = f (ln CC) forecast equation.

On the basis of the results obtained, it seems possible that the Coulter Counter values may correspond to some HIAC given values and vice versa.

Table 3 reports the mean and median forecast values, given the limit values set by the British and Italian Pharmacopoeias, as well as the intermediate values obtained by extrapolation, assuming that the cumulative number of particles–particle diameter distribution is log–log.

Table 3. Mean and median values forecast for HIAC and Coulter Counter according to the different size thresholds, based on present official limits.

	D (μm)	Coulter Counter (official limits)	HIAC[a] (forecast values) mean	median	HIAC (official limits)	Coulter Counter[b] (forecast values) mean	median
BP	2	1000	189	142	500	1707	1365
	3·5	245*	70	46	163*	564	342
	5	100	52	24	80	132	59
FUI	2	—	—	—	839*	2502	2000
	3·5	—	—	—	229*	712	432
	5	—	—	—	100	148	66

* Values obtained by extrapolation from official limits assuming a number of particles–particle size log–log distribution.
[a] Forecast equation HIAC = f (CC) was calculated on 37 batches.
[b] Forecast equation CC = f (HIAC) was calculated on 40 batches.

Finally, if we consider the correlation between the ln (limit values) for the different diameters and the corresponding ln (mean and median forecast values), in agreement with the literature, we observe a fairly good linear relationship for fitted ln HIAC (r_{mean} = 0·9967 and r_{med} = 0·9998) and for fitted ln CC (r_{mean} = 0·9868 and r_{med} = 0·9997), which would further confirm the reliability of the forecast values, even though these should be further verified with other series of data.

Evaluation of the British and Italian Pharmacopoeias' acceptability criteria

When analysed with the Coulter Counter method, 9 out of 40 batches (1, 2, 7, 12, 15, 16, 24, 32, 37) failed according to the BP criteria, but when the HIAC method was used, only two batches (26, 37) were unacceptable. This demonstrates that the limits set for the HIAC method are broader than those for the Coulter Counter method, also shown in Table 1.

The maximum contamination values set by the BP for the HIAC counter are much higher than those yielded by the statistical analysis of our experimental data, namely 500 particles ml^{-1} vs 189 or 142 for diameters exceeding 2 μm and 80 vs 52 or 24 particles ml^{-1} for diameters exceeding 5 μm.

The Italian official limits are higher than those of the BP for the HIAC method. On the other hand, according to the Italian requirements, the same batches would prove unacceptable (26–37) as those unacceptable according to the BP HIAC limits. This shows that, as far as the HIAC method is concerned, the acceptability criteria of the two Pharmacopoeias may be similar.

CONCLUSIONS

The results obtained allow several remarks to be made on the number of particles–particle size distribution pattern. The straight lines representing such distribution, as obtained for each solution with the Coulter Counter and the HIAC methods have a variable pattern, which is not typical of the type of solution examined. In many cases the two lines do not cross-over within the examined size range (2–25 μm) while in other cases they do cross, but at different size levels.

In general it may be concluded that the contamination values obtained with the Coulter Counter method are usually higher than those obtained with the HIAC method for sizes ranging between ≥ 2 and ≥ 5 μm. At higher size levels (≥ 10, ≥ 20, ≥ 25 μm) no general rule could be established.

The statistical analysis of results further clarified the relationships between the contamination values obtained with the two counting methods. For sizes ranging between ≥ 2 and ≥ 5 μm a relationship was found that allowed the forecast of values that could be obtained with the Coulter Counter method, based on the corresponding values obtained with the HIAC counter, and vice versa. The relationship decreases for diameters $\geq 3{\cdot}5$, ≥ 2 and ≥ 5 μm, while no

relationship could be demonstrated for sizes ≥ 10 μm.

On the basis of the maximum values set by the 1980 BP for the Coulter Counter method, the corresponding mean and median values forecast for the HIAC counter was calculated, and vice versa. Table 4 reports the suitably rounded values proposed for the Coulter Counter and HIAC methods, corresponding to the limit values set by the British and Italian limits for particles ≥2 and ≥ 5 μm in size.

Table 4. Values proposed for HIAC and Coulter Counter according to the different size thresholds, based on present official limits.

	μm	Coulter Counter	HIAC
		(official limits) ⟶	(proposed values)
	>2	1000	190
	>5	100	50
BP			
		(proposed values) ⟵	(official limits)
	>2	1700	500
	>5	130	80
		(proposed values) ⟵	(official limits)
FUI	≥5	150	100

When comparison of the number of batches acceptable according to the BP–HIAC and BP–Coulter Counter methods is made, and considering the values reported in Tables 3 and 4, it is evident that the limits set by the BP for the HIAC method are more permissive than those set for the Coulter Counter method, and therefore do not allow a correct evaluation of the acceptability of parenteral products.

The acceptability criterion set by the Italian Pharmacopoeia was found to be similar to the BP HIAC one.

The choice of the official particle contamination limits should take into account the instrumental method used. Theoretically, such limits should be defined so as to assure the same criterion of acceptability or, at least, that best practically definable. Table 4 lists the pairs of Coulter Counter and HIAC limit values allowing such a goal to be reached, with regard to the solutions examined.

Since it is known (Stembal 1983) that there is instrument to instrument variability (particularly for HIACs), recommendations for different limit values would need to be checked, not only for different parenterals but also for various instruments of each type.

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