EVALUATING GLASS, POLYSTYRENE, AND POLYPROPYLENE CONTAINERS FOR SEMEN COLLECTION AND SPERM WASHING

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The effects of glass, polystyrene, and polypropylene containers on semen parameters as measured by Cell Soft computer-assisted semen analysis and hypoosmotic swelling (HOS) tests were assessed in unwashed specimens and again after sperm washing and swim-up procedures. A superiority in unwashed specimens was observed in glass versus polystyrene concerning velocity, motility percentage, and HOS testing ($p < 0.01$). Also, superiority of glass versus polystyrene was demonstrated for linearity and ALH ($p < 0.05$). Glass was only superior to polypropylene in categories of motility and HOS testing ($p < 0.05$ and $p < 0.01$, respectively). There was no category in which plastic was statistically superior to glass in the unwashed specimens. Concerning washed and swim-up specimens, there were no semen parameters in which there was any superiority demonstrated of either glass or plastic. Unless some future proof that the differences in semen parameters demonstrated in this study have no clinical significance, these data suggest that the collection of semen samples for sperm analysis or therapeutic use should be performed in glass containers.

Key Words: Sperm; Semen; Polypropylene, Washing.

INTRODUCTION

Containers made of glass or different types of plastic have been used for semen collections. Calamera [2] found no difference in sperm motility or in fructolysis in plastic containers versus glass flasks. Balerna et al. [1] concluded that containers made of polypropylene or polystyrene were the best vehicles to achieve medium- or long-time conservation of human semen as judged by motility, PH, and oxidation.

A study was designed to further evaluate the effect of plastic versus glass on sperm parameters as measured by the hypoosmotic swelling (HOS) test and cell-soft computer-assisted semen analysis. Furthermore, these parameters were also evaluated after sperm washing and swim-up procedures.

MATERIALS AND METHODS

An equal aliquot of mixed semen specimens was divided into glass, polystyrene, and polypropylene containers from 20 specimens from normal donors. Additionally, 10 semen specimens were divided into

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TABLE 1 Sperm Parameters (Mean) in Glass, Polypropylene, and Polystyrene

Treatment	Velocity (μm/s)	Linearity	ALH Mean (μm)	CT $\times 10^6$ (Per ml)	Motility (%)	HOS (%)
Unwashed (n = 20)						
Glass	39.245	5.425	2.16	115.6	59.15	75.0
Polypropylene	37.685	5.165	2.11	118.4	45.95	64.4
Polystyrene	34.725	4.82	1.89	135.15	44.7	63.15
Washed (n = 10)						
Glass	42.39	5.43	2.181	128.5	59.4	74.3
Polypropylene	41.61	5.46	2.184	114.6	58.3	72.8
Polystyrene	43.23	5.36	2.2	132.4	56.8	70.6
Swim-up (n = 10)						
Glass	55.87	6.69	2.327	140.8	89.7	74.7
Polypropylene	56.03	6.48	2.418	134.3	85.4	70.8
Polystyrene	56.8	6.59	2.311	137.9	83.9	69.9

TABLE 2 T Values

Treatment	Velocity (μm/s)	Linearity	ALH Mean (μm)	CT $\times 10^6$ (Per ml)	Motility (%)	HOS (%)
Unwashed (n = 20)						
G *v* PP	0.943	1.070	0.401	0.148	2.172[b]	3.817[a]
G *v* PS	2.968[a]	2.227[b]	2.325[b]	0.907	2.672[a]	4.391[a]
PP *v* PS	1.663	1.181	1.561	0.793	0.228	0.418
Washed (n = 10)						
G *v* PP	0.427	0.091	0.018	0.579	0.136	0.436
G *v* PS	0.401	0.221	0.092	0.160	0.351	1.096
PP *v* PS	0.745	0.292	0.077	0.782	0.198	0.645
Swim-up (n = 10)						
G *v* PP	0.054	0.665	0.631	0.215	1.268	1.620
G *v* PS	0.306	0.326	0.122	0.092	1.650	1.961
PP *v* PS	0.225	0.337	0.904	0.148	0.416	0.426

[a]Significance 0.01.
[b]Significance 0.05.

glass, polystyrene and polypropylene centrifuge tubes, and washing and swim-up procedures were performed [3]. The unwashed semen specimens were incubated at 37 °C, 5% CO_2 for 1 h, after which time they were evaluated by Cell Soft computer-assisted semen analysis and HOS test. The HOS test was performed as previously described by Jeyendran et al. [4]. The washed sperm samples were analyzed after 1 h incubation in Ham's F-10 media.

RESULTS

The results can be seen in Table 1. In unwashed specimens sperm velocities were better in glass versus polystyrene ($p < 0.01$). Although the velocities were better in glass than polypropylene, the difference was not significant (Table 2). Also, there was no significant difference in polystyrene versus polypropylene.

The same comparison held true for measurements of linearity and ALH. There was only significance in the difference between specimens contained in glass versus polystyrene (both $p < 0.05$). The different materials did not affect the sperm count.

In unwashed samples motility was significantly greater in glass than in either polypropylene ($p < 0.05$) or polystyrene ($p < 0.01$). The difference was even greater when evaluating the HOS test; glass gave higher values than polypropylene or polystyrene ($p < 0.01$).

For 10 washed and 10 swim-up specimens there were no significant differences in any of the sperm parameters in different material containers.

DISCUSSION

Although some previous articles have suggested that polypropylene and polystyrene do not exert any negative effect on sperm function parameters, the present data indicate a definite superiority of glass containers for unwashed specimens, using more subtle measurements of velocity, linearity, ALH, and HOS testing.

The reduced motility would be most likely secondary to interaction with chemicals from the plastic. Strickland and Ziaya [5] similarly found a decrease in motility in semen samples placed in sterile plastic urine cups. By using a computer type of analysis, possible judgment bias by the technician is eliminated by this study. Also, the decrease in HOS test might be important because of some data that correlate this test with in vitro fertilization outcome [6].

Whether these differences would have any clinical adverse effect is not known. However, unless some future proof that the differences in semen parameters demonstrated in this study have no clinical significance, these data suggest that the collection of semen samples for sperm analysis or therapeutic use should be performed in glass containers.

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