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ORIGINAL PAPER

Comparative amperometry on ocular-surgery electrolytes in eye-splinter trauma from galvanized-steel electrochemistry

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Abstract Metal foreign bodies in the eye are commonly removed utilizing a surgical approach through the vitreous cavity, which requires a maintained ocular pressure by infusion of replacement "irrigation solution", i.e., electrolyte. However, when in contact with aerated electrolyte, if the fragment comprises galvanized steel, the component metals can act as a bimetallic zinc/steel cell, thereby electrochemically generating injurious amounts of hydroxyl ion OH^-. The effect on this electrochemical reaction of the compositions of the irrigation solutions in use, and of their analogues, has thus been investigated, to seek the least damaging. *The inter-metallic current relates directly to the rate of the injurious reaction*, hence amperometry has been the principal probe employed in our study. Establishing reproducible conditions of contact between electrolyte and the steel–zinc combination forms a major focus of the study, which is to attain amperometric reproducibility in the comparison of the electrolytes. Geometry-sensitive directed injection of test solution into a cell with pre-fixed separate steel and Zn electrodes achieved the desiderata. Besides potentiometry and amperometry, pH and buffering by the various test solutions, and the effects of surfactants, were also examined. Free-radical intermediates in the oxygen processes are further injurious suspects. Anaerobic conditions that would preclude both intermediates and OH^- are unfortunately damaging to the structures of the eye, so alternative surgical procedures are needed. Anti-corrosion aspects are considered.

Keywords Vitrectomy solutions · Intraocular foreign body · Amperometry · Galvanized · Zinc-steel · Ocular trauma · Oxygen · Hydroxyl · Anti-corrosion

1 Introduction

When eye trauma surgery overlaps corrosion studies, basic expositions, experimental compromises, and some new approaches are called for. To successfully remove a metal splinter from an eye, the vitreous in the eye must be excised with a fast-cutting *vitrectomy machine*. As the vitreous is removed via an aspiration port on the vitreous cutter, an electrolyte solution [an 'irrigation' or 'vitrectomy' solution, necessarily air-saturated] replaces the lost vitreous to maintain equal pressure in the eye throughout the operation. Many single-metal splinters (or intra-ocular foreign bodies, IOFBs) can be successfully removed in this manner, but a galvanized (i.e., zinc-coated) steel fragment, in contact with electrolyte, drives electrons 'e' from the zinc to the steel areas, producing OH^- from the oxygen in the electrolyte. Reaction of this generated OH^- with the retina has been deemed to cause at least partial blindness [1]. The electrochemistry, well known in anti-corrosion studies [2], is summarized as follows.

$$\mathrm{Zn(s)} \rightarrow \mathrm{Zn^{2+}(aq)} + 2\mathrm{e_{Zn}} \quad \text{(spontaneous Zn dissolution)} \tag{1}$$

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$$2e_{Zn} \rightarrow 2e_{Steel} \quad \text{(inter-metal e transfer by metallic conduction)} \tag{2}$$

$$\tfrac{1}{2}O_2(aq) + H_2O + 2e_{Steel} \rightarrow 2OH^-(aq) \quad \text{(on steel)} \tag{3}$$

$$\text{Overall } Zn(s) + \tfrac{1}{2}O_2(aq) + H_2O \rightarrow Zn^{2+}(aq) + 2OH^-(aq) \quad \text{(sum of (1) to (3))} \tag{4}$$

Various vitrectomy solutions are in use, or contemplated. Thus, it is important to compare these solutions, when in contact with Zn and steel, for the rapidity of the noxious electrochemical reaction, so as to employ that showing the slowest reaction, which would be the least injurious. *This reaction rate is directly proportional to the rate at which electrons transfer between the Zn and the steel, and may be measured by separating the two metals in order to meter the intermetallic current externally across a suitable cell,*

$$Zn \mid \text{test solution} \mid Steel \tag{5}$$

Separation of the two metals to act as electrodes in a cell requires rewriting reaction (2) as

$$e_{Zn} \rightarrow e_{Cable} \rightarrow \text{meter} \rightarrow e_{Cable} \rightarrow e_{Steel} \tag{2'}$$

so allowing amperometry, or potentiometry, of the relevant electron flux arising with each test solution.

The experimental challenge has been to simulate, in the laboratory cell, the highly chaotic conditions at the moment of contact between the test solutions and the metals, approximately as they might occur within the eye in surgical operations. In the present comparative study, the immediate external Zn-to-steel current is measured on contact of the zinc and steel electrodes with each vitrectomy solution, or their alternatives.

Furthermore, from these observations, some reaction-inhibiting additives are explored, other possible injurious species (progenitors to OH^-) are noted, and remedial action is discussed.

2 Materials

2.1 Solutions

All the test solutions are "isotonic" or nearly so, that is, of the same osmotic pressure as body fluids. All the solutions, though syringed from polythene sacs or auto-resealing bottles, are saturated with O_2 and CO_2 in use and in the tests. They are listed here with the suppliers' statements of the compositions, which are not always complete. Labels A to F allow of concise text reference.

[Our comments are included below in square brackets. An oft-used abbreviation for the solutions in use is "% w/v", for mass g dissolved in 100 cm^3 of solvent. $M = \text{mol dm}^{-3}$.]

Solution A is Bausch & Lomb's "Aqsia", 'Balanced Salt Solution for ocular use': of composition $CaCl_2 \times 2H_2O$; $MgCl_2 \times 6H_2O$; $C_2H_3NaO_2 \times 3H_20$ [? i.e., CH_3COONa?]; NaCl; KCl; H_2O [No concentrations are given, and the unexplained "×" is probably unnecessary].

Solution B is Freeflex "Hartmann's solution", code 2743531: 'Compound sodium lactate' K 5 mM, Na 131 mM, Ca 2 mM, Cl [chloride] 111 mM; bicarbonate (as lactate) [*sic*] 29 mM.

Solution C is Macoflex Sodium Chloride 0.91% w/v: [i.e., NaCl 9.1 g dm^{-3} aqueous, which is ~0.15 M, approximately isotonic].

Solution D is Macoflex: Sodium Chloride 0.18% w/v, Glucose 4% w/v.

Solution E is Macoflex Glucose 5% w/v; pH 4.15.

Liquid F is Elga laboratory de-ionized water [adventitious impurities, e.g., from glass, and dissolved CO_2, are ca. 10^{-5}–10^{-6} M].

2.2 Surfactants as solutes

For the study of surfactant effects, sodium hyaluronate (or 'Hyaluranon') [3–6][1], sodium chondroitin [7] (*ex* shark cartilage), and chitosan [8, 9] *ex crustaceae*, were all from Sigma, and the *n*-hexadecyl-1-trimethylammonium bromide (or HTMA) was from Aldrich.

2.3 Electrodes

Pure zinc (99.9%) from BDH/Merck was used in preference to spectrographically pure Zn, being more like galvanizing Zn (in prior experience [10]). From a strip cut from a ~0.15-mm-thick sheet, a face of 1.42 × 0.61 cm^2 was left rectangular when the remaining ~1.5 cm length was rolled to form a ~1-mm diameter cylindrical tube for protruding through the cell lid.

For the steel electrode, a mild steel rod 6.5 mm in diameter was machined down at one end to give a (lengthwise) rectangular face of 1.47 × 0.62 cm^2. Non-electrode areas were insulated with varnish and only the tops protruding through the cell lid (described below) were partly clear for external electrical connections. These projecting extensions were fixed in place externally with silicone rubber cement to give a 0.8-cm electrode separation within the cell.

Both the Zn and the steel electrode areas were initially abraded with Oakes glass paper BS 871 Grade S2 Grit 40,

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[1] The first hyaluronan biomedical product… is approved for use in eye surgery (i.e., corneal transplantation, cataract surgery, glaucoma surgery and surgery to repair retinal detachment) [and for]… other ophthalmic surgery.

followed by Halford's finest-grade aluminum oxide paper P1500, each time for 15 s. Obvious striations from the glass paper abrasions were thereby smoothed to a shiny finish. Finally, each surface was gently wiped with tissue paper for 8 s to remove the abraded dust.

The state of the cell electrode surfaces could only approximate the ocular conditions, but in the lab, pre-experimental treatment was important for reproducibility: abrasion sequences as detailed above gave near-mirror finishes, but obviously striations at > nm scales will have persisted. Pure metal surfaces were never accessible anyway [also of course during the occurrence of an ocular injury] as, within microseconds, hundreds or even thousands of oxide layers spontaneously form in air [11]; once formed we believed them to be quite stable.

3 Experimental: preliminary electrochemistry

With the two electrodes connected, when they were immersed in phenolphthalein-containing oxygenated electrolyte, indicator coloration on the steel indicated rapid generation of OH^-, but in a separate test on Zn alone, indicator experiments show that abraded [i.e., shiny and almost oxide-free] Zn surfaces also liberate OH^- from oxygen by the direct chemical reaction represented by reaction (4) above, much more slowly however than via the electrochemical process, as expected [2]. Other metals, which ordinarily show atmospheric corrosion, could similarly produce OH^- by direct reaction, which must be of ophthalmological concern.

In preliminary experiments, Zn and steel electrodes suspended in a fixed fashion from a lid were plunged into the test solutions. Highly approximate cell emfs E were thus observed an arbitrary 0.2 s after contact, the delay necessarily obviating the infinite values predicated by the Nernst equation for zero product concentrations (1 mM Zn^{2+} added to each solution gave similar data). However, the *more important current measurements* in these trial 'plunge' experiments were irreproducible, with successive values varying by as much as 40 or 50%. Hence a detailed study, of the outcome of many trials, is given below, as they led to *a different experimental procedure ensuring satisfactory reproducibility*.

To avoid the irreproducibility of currents, preliminarily evident in the "plunge" experiments, solution injection into a cell with lid-supported electrodes rigidly in place was studied in detail. Thus, injection upwards from the cell base, or downwards through the lid, or laterally halfway up the side of the cell, all proved unsatisfactory. Reproducibility was only achieved as described below under "*…contact procedure.*"

4 Experimental: the method achieving reproducible amperometry—the cell vessel and the contact procedure

The vessel comprised a polythene cylinder 2.8 cm high and 1.1 cm i.d. An external 'cap', comprising an inverted cylindrical container 5 mm high, fitting snugly outside the top of the cell vessel, acted as electrode-supporting lid. An injection hole (1.5 mm diameter) in the side of the cell vessel was positioned so that with the cell assembled, liquid could be squirted in between the 'shoulders' of the electrodes. A 50-cm^3 syringe was used via its snub nose inserted into the injection port to manually inject 2.0 cm^3 of test solution within about 0.1 s. The syringe, held at ca. 20° to the horizontal, then filled the cell with least turbulence and no 'splash' loss via the lid, which had a small 'exhaust' tube for expulsion of the displaced air. While mechanical devices to drive injection were contemplated, the need to abrade the electrodes before each measurement required the rapid dismantling possible with our arrangement. Solutions were always exposed to air at 24 ± 1°C for >15 min before amperometry.

4.1 Amperometry

Original measurements recorded on an oscilloscope were initially too chaotic from turbulence to be useful, and a pen recorder [Lloyd 'Graphic' model 450] proved better, via its mechanical damping of extreme excursions, over the initial 0.1 s. The currents generated by the cell were passed through a 1-Ohm standard resistance [a value far less than the intrinsic cell resistances] across which the potentials were recorded for amperometry. The effects of adding the surfactants, listed in 'Materials', on the amperometry of some electrolytes, were also studied.

5 Results and discussion

The prescribed amperometric procedures need to be closely followed for reproducibility of currents within a few percent. While such data, somewhat dependent on cell geometry, are clearly not fundamental, the *relative* responses of the different test solutions will be reproduced by similar experimentation, even if any set of *observed* values differ numerically from ours. It is the *relative* responses of the different test solutions that comprise the objective here. Clearly, the ideal response would be zero current, hence zero eye damage, but *faute de mieux* the least current observed would pick out the most desirable irrigation solution. The basic electrochemistry [2] can be described as 'mature' and we had no intention to elaborate on it, e.g., by individual-electrode cyclo-voltammetric

Table 1 Amperometry on successive samples of each test solution, exemplifying reproducibility

Solution	Currents (mA)	Averages with averaged |deviation|
A	23.0, 20.6, 21.0, 23.5	22.0 ± 1.2
B	22.5, 21.5, 22.5, 22.7, 22.0	22.2 ± 0.4
C	22.3, 25.0, 25.0, 24.95	24.1 ± 1.1
D	11.5, 12.5, 11.0, 10.5	11.4 ± 0.4
E	0.255, 0.205, 0.190, 0.220	0.22 ± 0.02

studies. We note only that the slower reaction is the multistep O_2 to OH^- reaction on most metals, cf. the facile Zn-ionizing dissolution generating electrons [2].

Because the experimental amperometric approach that we arrived at is highly empirical, and was initially poorly reproducible especially in the 'plunge' experiments, individual data in separate measurements are detailed in Table 1 to demonstrate the eminently satisfactory reproducibility ultimately attained as described above, following our various solution-injection approaches. To repeat, there is no 'ideal' current or limit: any irrigation solution showing generation of a current markedly lower than any other would be the desirable candidate for surgery.

Other test-solution data together with the mean currents are given in Table 2. Solutions A and B, Bausch and Lomb "Balanced Salt Solution" and "Hartmann's Solution" respectively, are commonly used in vitrectomy. Solutions A to C (Table 2) all give similar currents and quite close values of the observed cell potentials E, but solution A does give an E value >0.6 V cf. ≈0.50–0.55 V for solutions B and C. Thus, the current data do not pick out the least-damaging electrolyte from present availabilities for ocular surgery. Solution D produces a lesser current doubtless owing to the lower concentration of electrolyte, but ophthalmologists disfavor its untested use. This applies even more strongly to electrolyte-free solution E (isotonic glucose solution), which is expected to damage ocular components. Solution E, and the lab de-ionized water F, show comparable amperometry, as expected.

Table 2 Cell potentials and currents from vitrectomy and other solutions, with initial pH measurements, and also the slightly-alkalized pH values

Solution	Potential[a] (V)	Current[b] (mA)	pH[c]	pH(alk)[d]
A	0.78–0.65	22.0 ± 1.2	7.02	7.20
B	0.61–0.50	22.2 ± 0.4	6.08	6.42
C	0.63–0.51	24.1 + 1.1	5.5	6.5–7.1
D	0.62–0.57	11.4 ± 0.4	Untested	
E	0.62–0.50	0.22 ± 0.02	Untested	
F	0.53–0.47	Variable, <0.2	7.2 ± 0.8	9.4 ± 0.3

[a] From 'plunge' experiments, where the initial (0.2 s delay) values decreased on product accumulation following the Nernst eqn.; the range observed is given

[b] Fixed-electrode injection measurements as in "Experimental"; detailed data in Table 1

[c] pH of D and E unreliable owing to adventitious oxidation-products of the glucose component

[d] After 0.1 cm^3 addition of 0.02 M NaOH to 20 cm^3 of test solution. Bulk measurements

5.1 pH, buffer action, and surfactants

Thus, apart from suggesting some mitigation of injury by the use of solution D, the amperometric tests might seem disappointing, but do point to possible chemical remedies (below). The concentration of O_2 in water and the eye [12] is ~2 mM, giving 8 mM as an upper limit to OH^- from "total" electrochemistry, but this maximum is unlikely within the eye. Approximate coulometry shows the electrogenerated OH^- to give a pH of ~9 in our experimental cell and possibly up to 10 about an ocular splinter. The pH values of the test solutions were measured (Table 2)and then an amount of OH^- calculated to confer a pH of 10 on water was added to each of these solutions, giving the "pH(alk)" values in Table 2. Both solutions A and B show considerable buffering with changes of ~0.4 or less in pH, while the unbuffered NaCl Solution C shows on alkali addition a greater shift, moderated however by its initial slight acidity [undeclared by suppliers re our sample]. In contrast, the Elga deionized water approaches the expected value of 10. Despite the noted buffering by the widely used irrigation solutions A and B, ocular trauma in surgery still occurs, and if OH^- remains the culprit, the trauma may set in before its interaction with the buffer. See *Wider Chemical Considerations* below.

For counter-measures, some electrochemical considerations apply: the inclusion in a vitrectomy solution of a charged surfactant, preferably biochemical, could inhibit the reduction of O_2 by attachment to one or other metal surface, as part of the double layer that must form on electrodes during incipient current production. Surfactants commonly act at low concentrations. The charged head of an anionic surfactant would seek the positive surface of Zn (which loses electrons) while that of a cationic surfactant will be drawn to the negative steel surface charged by electron acquisition. Negatively charged chains of Na hyaluronate or "hyaluronan" [3] noted as being ophthalmologically useful [6][2], when added to electrolyte, proved ineffective as a reaction inhibitor, merely increasing the current on addition. The results appear in Table 3 where individual current measurements are given [again to demonstrate acceptable reproducibility], with averages

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[2] See foot note 1.

Table 3 Surfactant addition (tests 1 and 2, anionic surfactants; tests 3 and 4, cationic surfactants)

Test no.	Electrolyte	Current (mA)	Surfactant added	Current (mA)
1	$ZnCl_2$(aq), 0.0046% w/v	1.85	Na hyaluronate	2.95
		2.40	0.091% w/v	2.95
		1.75		2.95
		1.35		3.00
		1.85		Av. 2.96 ± 0.02
		Av. 1.84 ± 0.23		
2	NaCl(aq), 0.03% w/v	5.2	Chondroitin	6.90
		6.35	0.09% w/v	6.35
		5.90		5.90
		Av. 5.82 ± 0.41		6.05
				5.35
				Av. 6.11 ± 0.41
2a	None		Chondroitin	1.55
			0.09% w/v	2.10
				Av. 1.82
3[a]	NaCl(aq), 0.09% w/v	22.0	Chitosan	23.5
	made pH 5 with HCl	23.0	0.05% w/v (no HCl)	23.5
		23.5		22.5
		21.5		24.0
		Av. 22.5 ± 0.7		Av. 23.4 ± 0.4
4	NaCl(aq), 0.09% w/v	19.5	HTMA	13.5
		19.5	0.1% w/v	18.5
		19.3		14.0
		19.0		10.0
		Av. 19.3 ± 0.2		Av. 14.0 ± 2.3
4a	None		HTMA(aq)	0.55
			0.1% w/v	0.68
				Av. 0.61

[a] Note to test 3: Chitosan alone in H_2O became saturated at 0.05% w/v and showed a pH of 5

(“Av”) ± averaged deviation. The anionic surfactant sodium chondroitin [7] also failed (test 2, Table 3), its addition again just enhancing the current. A saturated solution of chitosan alone showed slight acidity; thus the electrolyte was acidified to the same pH for its (solo) amperometry, but not in its mixture with the chitosan (test 3); this procedure ensured the similarity of conditions. Notably, addition of chitosan did *not* enhance the current.

By contrast, the common surfactant HTMA, *n*-hexadecyl-1-trimethylammonium bromide, provided an unambiguous decrease in current (test 4, Table 3), with initially puzzling scatter, intelligible however by considering details of the steel surface. Nanometer striations from the pre-abrasions, because of their averaging-out, will be irrelevant to gross areas governing small-molecule or monatomic-ion electrochemistry (reported in Table 2) as in reactions (1) and (3). However, in successive experiments with attached surfactant like HTMA, the detailed variation in the striations will provide varied chain orientations to the HTMA specific to each experiment. Multi-directional surfactant orientations, the heads adsorbed as inner part of the double layer, will be set by these striation geometries, with ensuing variability between experiments in allowing access to the approaching O_2(aq) molecules about to undergo reduction. There is accordingly an exceptional variability in the currents (cf. the surfactant-free data, Table 3). Only positively headed surfactants will be adsorbed (in double layers) on steel and show this effect.

Negatively headed surfactants, necessarily linked to Zn not steel surfaces, might somewhat hinder the release of Zn^{2+} from the metal in reaction (1), but no effects are shown in tests 1 and 2 of Table 3 (Hyaluronan, bearing charges along its chain, might have been expected to be more effective here).

Ocular tolerance in surgery to higher concentrations of HTMA (to more strongly inhibit the O_2 reaction) is problematic. However, a 2+ surfactant employing a Cr(III)-headed chain, such as $CH_3(CH_2)_nCOOCr^{2+}$, should certainly outperform the mono-positive HTMA. [Cr is suggested here because Cr^{III} is almost uniquely retentive of its ligands]. Such an additive would also need to be ocular-inert. Without diminishing the current, chitosan did not enhance it, which might indicate a feeble, marginal, inhibition. Compared to HTMA, its bulky structure could result in a sterically limited surface interaction that diminished its inhibitory efficiency.

5.2 Wider chemical considerations

While the close comparability of current magnitudes observed for vitrectomy solutions precludes choice of any particular one against electrochemical trauma, other factors may apply. Thus even if final OH^- production is suppressed possibly by buffer action, fleeting intermediate species arising in its formation could still inflict injury. Thus, O_2 gets reduced through the formation of hydrogen peroxide H_2O_2, formed via the free radicals [13] $HO_2^\bullet$ and $O_2^{-\bullet}$, which are highly destructive species.

Furthermore, Fe^{2+} if present reacts [14] with H_2O_2 to give Fe^{3+}, $HO^\bullet$ and OH^-. The free radical is especially reactive. Appropriate surfactant in a vitrectomy solution may offer protection against these species.

De-aeration of the irrigation solutions, and anaerobic surgical conditions, seem to offer a remedy, but eye components, being avascular, need direct oxygen, otherwise will suffer severe and rapid degradation. However, provided the galvanized splinter or IOFB can be kept away from the retina macula by appropriate approach surgery (“tunnel vitrectomy”), damage could be minimized by being largely confined to the vitreous [15].

Zinc/steel IOFB compositions are established in post-operative analyses, but surgeons benefit from information regarding the source of the fragment: examples hitherto have been (1) a galvanized garden-wire offcut impelled during mowing, (2) a galvanized fragment hammered from a tractor attachment, and (3) a galvanized nail being hammered into concrete.

6 Conclusions

In detailed studies of the cell (5) above, a successful amperometric protocol of acceptable reproducibility was ultimately arrived at, which could find wider usage. The cell was used to compare the various vitrectomy solutions and their analogues. The data showed closely identical currents with the different vitrectomy solutions at present in use, all evoking similar reaction rates of OH^- production from the galvanized steel systems. Lower currents (Table 2) were seen only with solutions deemed unfavorable for vitrectomy. So an investigation was undertaken into possible surfactant solutes that would become attached to electrode surfaces as components of the double layers on the electrodes. This showed at least one species, the monocationic HTMA, to effect an appreciable decrease in the injurious reaction as far as can be judged from these lab experiments. More highly charged cationic surfactants would undoubtedly be more effective. When possible, however, a different surgical approach could preserve the retina from reaction damage [15].

In technology, a much-used galvanized surface could need painting for continued protection, and from our studies, a possible anti-corrosive inhibitory effect of a suitable surfactant component in the coating merits consideration.

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