Preparation of a Rechargeable Battery Using Waste Protein from the Fish Scales

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Summary: The electrochemical redox reactions of the oxytocin and fish scale protein which are mainly collagen were exploited for the preparation of a rechargeable protein battery named as fish scale battery. This battery was found to depend upon the concentration of oxidizing and reducing agents, voltage of the charger and the time for charging. Some of these parameters were optimized using a single cell of this battery and some others were optimized by using five cell battery. The five cell protein battery gives a maximum and stable voltage of 8500 millivolt. The way of charging and theoretical aspects of the battery is also discussed in this communication.

Keywords: Fish scale battery, Oxytocin anodic half, Electro-active collagen, Collagen redox, Peptide analysis

Introduction

Proteins, peptides and amino acids are electrochemically active biomolecules due to their polarity and ion accepting properties. The electrochemical reactions of these compounds also used in medicines, analysis, electricity storage and waste monitoring and management [1-4]. One of the most important applications of the electrochemical activity of these compounds is theirs capacity to store the electricity. Our group introduced a rechargeable protein battery using the protein extracted from the chick pea and kidney beans [4]. The aim of current study is storage of electricity and management of waste protein. For this purpose we explored one of the most important sources of proteins *i.e.* fish and fish industries waste products. In view of its greater contribution in the food and other products its production increasing day by day. According to an estimate 131 million tons of fish was produced in 2000 which was increased to ≈160 million tons in 2006- 2007 [5,6] and almost 74% of this was processed for food [6]. Almost 40-70% of the material is discarded as waste [7] which contains viscera, shell (from crustacean and molluscan processing), scales, fins and frame bones [8]. More than 30% of fish processing wastes consist of skin, scale and bone [9]. Excessive amount of the fish waste causes environmental pollution in terms of the foul smell, emission of toxic gases and pathogenic microbes [10,11]. It may also cause water pollution and increase the biochemical oxygen demand (BOD) which is a threat to the aquatic life [5]. Various workers introduced various methods for the disposal of the fish byproducts from processing and distribution units. These methods include the extraction of proteins, amino acids, food supplements and oil from the waste in addition to its use for fertilizer. Some of the waste can also be fed to the animals [1,8,9,12-16]. Among the byproducts of fish industry scales are very hard to decompose and none of the animals use to eat them. Their chemical composition for various species of the fish indicates that these are mainly composed of collagen and very small quantity of carbohydrates in addition to minerals [15]. Various researchers reported the extraction and characterization of collagen from the fish scales [8,9,12,14]. The fish scales were also reported to be used for the adsorption of dyes, metals and also for enzyme based electrochemical and fluorescence analysis [2,3,13,17].

The present work is aimed to utilize the fish scales for the preparation of rechargeable battery by coupling it with oxytocin, a low cost and easily available peptide. The idea is based on the electrochemical properties of the collagen and oxytocin. This battery may not only be utilized for storing the electricity but may also help in the evaluation of the electrochemical properties of the collagen and the utilization of this waste for useful purposes.

The basic theory of protein battery can be described on the oxidation and reduction reactions of proteins, peptides and amino acids. The relative

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affinity of these biological compounds towards the redox reactions is different due to which these have difference in electrode potentials. These activities may be due to the oxidation of these with oxygen or some polymerization reaction like the sulfur bridge formation. Literature reports indicate that these compounds may pass through electrochemical oxidation under appropriate conditions [2,3]. The electrochemical oxidation and reduction of the proteins and peptides may be a reversible reaction due to which oxidized and reduced forms of these compounds may remain in equilibrium. Coupling of proteins or peptides with each other in a cell through a salt bridge may form an electrochemical cell.

Experimental

Material and Method

Fish scales were collected from a fish farm/shop in the Mardan City of Khyber Pakhtunkhawa Pakistan through a random collection. Three types of the fish were sold and processed by that shop; the Rohu, Grass carp and Catla. The scales were predominantly composed of Grass carp. The scales were properly cleaned from the blood using tape water and then distilled water and were dried in the hood at the ambient temperature. Clean and dry fish scales (250 g) were dissolved by heating in enough quantity of the 3% aqueous solution of sodium hydroxide to obtain a final solution of one liter. This solution was named as fish stock and used as the cathodic half of the battery. The concentration of this solution was reported in terms of the concentration of sodium hydroxide in optimization reactions. For the anodic half of this battery, oxytocin was used as electro-active species. For this study pharmaceutical grade oxytocin was obtained from the local market which was further diluted according to the experiments under consideration. Each time this solution was basified according to the basic strength of the other half. The container for the battery was composed of 5 units and each unit of the battery was prepared by joining two cubical using epoxy resins. Each of the containers has a volume of 3.2 cm^3, a cut was made in the joining walls for facilitating the salt bridge. Graphite electrodes for this were obtained from the dry cells. These electrodes were placed in a card board for the preparation of circuit which acts as supporting and insulating material. The connections were made through copper wire according to the circuit requirements. All the anodic halves were filled with the aqueous solution of oxytocin and cathode halves were loaded with the aqueous solutions of fish dissolved by the aqueous solution of sodium hydroxide. The two solutions were connected by the salt bridge and the voltage was measured after placing electrodes in the solution. Electrodes of different material like graphite, copper, and lead were used for the investigation of the effect of electrode on the voltage of battery. The size of these will be described in the result and discussion part. The effect of salt bridge on the voltage of battery was investigated using different material as salt bridge. These were wick of the spirit lamp, lantern wick and a piece of card board. All these were obtained from the local market and used as salt bridge after proper cleaning with different solvents. The card board was soaked in an aqueous solution of the 0.1 M sodium hydroxide for 24 h which makes the salt bridge an electrolytic conductor. This cell can generate electricity according to the difference in electrode potential of the two halves and may be recharged. All the voltage measurements were carried out using digital multi-meter. The battery was charged using indigenously-prepared chargers of various strengths according to the procedure. Each experiment was carried out in triplicate and the average was reported (Fig. 1-4).

Fig. 1: Four cell protein battery.

Fig. 2: Strip of Electrodes.

Fig. 3: Single cell battery.

Fig. 4: Cotton wick for salt bridge.

Results and Discussion

Effect of Electrode on the Fish Scale Battery

In the fish scale battery, the fish scale protein was used as the reducing half and oxytocin as the oxidation half. The electrodes for this act as the conductor therefore, electrodes of the same nature were placed in each half. It was believed that the nature of electrode may play role in voltage of the battery, either due to the catalytic action of these for the oxidation or reduction of the proteins and peptides. In addition, it may be due to the active chemical reaction of the oxidized and reduced species with the electrodes under consideration which was investigated using four types of the electrode. The results of this study are given in Table-1. It can be seen from Table-1 that the voltage for aluminum and graphite are similar which may be due to the relative inertness of the two. In case of the lead it is far greater which may be due to the catalytic or chemical action of the lead. In case of copper the voltage is less than all which might be due to some retarding activity of the copper. Based on the inert nature of graphite it was selected as the electrode material for further work.

Effect of Salt Bridge on the Voltage and Stability of Fish Scales Battery

It was reported in our previous work on the protein battery that the nature of salt bridge also plays a role in the voltage and stability of the battery [4]. It connects the two solutions and may help to keep the free energy difference of the two solutions. In case of the more porous salt bridges the free energy difference decreases which lead to less stable voltages of smaller quantity and reversal is true for relatively less porous. Moreover, the use of salt bridges of very small number of pores is also non-beneficial due to lack of conductivity. The effect of the nature of salt bridge on the voltage and stability of the battery was investigated using a single cell. The cell was recharged for 20 minutes using a 12 volt indigenous but calibrated charger. Highly porous wick of spirit lamp, lantern wick made of woven threads of intermediate porosity and a less porous salt bridge of card board were used. Card board was used in two modes, one in the highly hydrated form and another in non-hydrated or less hydrated form. The results of this study are given in Table-1. If a salt bridge of a spirit lamp wick, composed of loose threads, was used the voltage and stability is less compared to the usage of a compact wick composed of waved wires as used in lanterns. In case of the use of a highly hydrated card board as a salt bridge, a further increase in stability and voltage was observed. But for less hydrated it was less than the lantern wick salt bridge. This is due to the less conductive nature of the non-hydrated salt bridge. Based on the greater value and more stable voltage, hydrated card board was selected for further work.

Table-1: Investigation of suitable salt bridge for the fish scales battery.

S. No.	Salt Bridge	Voltage (milli volt)
1	Spirit lam wick	113.0
2	Waved thread wick	140.0
3	Highly hydrated Card board	180.0
4	Less hydrated card board	80.0

Investigation of the power of charger on the charge storage capacity of the battery

The power of the charger may also play a role in the voltage of the fish scale battery. It was investigated using a five cell fish scale and oxytocin battery. The increase in power of the battery may change the voltage of battery by two possible mechanisms. Either may be increase in concentration of the oxidizable and reducible species or electrochemical formation of compounds which may have more capacity of the storage of the power. The results of this study are reported in Table-2. It can be seen from Table-2 that the voltage of battery increases with increase in power of charger. It seems that increase in the power may bring more increase. It supports the idea of the change in equilibrium constant. Although the voltage is greater in case of the 36 volt charger but we used 12 volt charger for most of the experiments.

Table-2: Investigation of the power of charger on the charge storage capacity of battery.

S. No.	Max voltage charger (volt)	Charging time	Voltage (milli volt)
1	12	20	6500
2	24	20	7400
3	36	20	8500

Investigation of the optimum time of charging for fish scales battery

Oxytocin is a peptide having the tendency of oxidation on coupling with proteins obtained from the fish scales. Both, oxidation and reduction half of this battery observes equilibrium phenomena. The equilibrium constant of which is a function of the input energy. Continuous supply of the electrical energy of DC (Direct current) type resulted in the equilibrium to shift toward the left. This increase in concentration of the reactant under the influence of the electrical power is responsible for the storage of power. An increase in the charging time of the battery may increase the amount of the oxidizable and reducible species which may either increase the voltage of the battery or the power of battery. Optimum time for the charging of battery was investigated using a battery composed of five cells. Each of the time optimization experiment was carried out in triplicate and the average was reported. This battery was charged by a 12 volt calibrated charger. It can be seen from the results in Table-3 that increase in the charging time of the battery increases the voltage of the battery until a limiting value. This limiting value of voltage indicates a limiting value of the concentration of the reducible and or oxidizable species. Based on the maximum voltage of the cell, 20 minutes charging time was selected for further work.

Table-3: Investigation of the optimum time of charging for the fish scales battery.

S. No.	Time / minute	Voltage (millivolt)
1	5.0	280
2	10.0	380
3	15.0	440
4	20.0	650
5	25.0	650

Investigation of the optimum amount of oxytocin and Fish scale solution

Electro-active species of the each half of this battery may pass through reversible reactions. Each half of this battery observes equilibrium between the oxidized and reduced species. In this case both the activity and total concentration of the electro-active species play role in the voltage, stability, and life/power of the battery. Compounds having antioxidant activity may be used as redox species with other species of the same activity. Various reports indicate the antioxidant activity of the peptides [18,19] therefore, we have used oxytocin as the anodic half of our battery which is sulfur containing peptide and can pass through reversible electrochemical transformations. This molecule contains various sites for redox reactions which include the sulfur linkage, oxygen moieties, hydroxyl and the amino groups. Literature reports indicate that OH containing compounds like phenols have antioxidant activities which may change according to the number and environment of the OH groups [20-22]. The cathodic half containing an aqueous alkaline solution may also contain peptides and amino acids due to the hydrolysis of the protein of fish scales which pass through electrochemical redox reaction. The concentration of the active species in both halves may depend upon the amount of the solutions which was investigated for each of the two in separate assays. Each time one of the reactant was kept constant and the other was varied. The volume of each solution in the half cell was 50 mL. However, the concentrations were different but the concentration of the base was the same in both half cells. The concentration of the fish scale solution was reported in terms of the concentration of the sodium hydroxide while that of oxytocin in part per million. The results of these studies are given in Table-4 and Table-5. Each time a single cell rather than battery was used for these investigations. The salt bridge of the cell was made-up of a piece of extensively hydrated card board and charger of 12 volt was used for recharging the cell. The cell was recharged for 20 minutes in each of the experiments. It can be

observed from Table-4 that the voltage of the cell varies with changes in the concentration of the electrochemically active proteins and peptides of the fish scale. Based on the maximum voltage 0.1 M solution of the fish stock was selected as the optimum concentration of the fish stock. The results for the variation of oxytocin are given in Table-5. It can be seen from the results that the voltage increases with the increase in concentration of oxytocin. A maximum voltage was obtained at 0.15 ppm concentration of the oxytocin and it was selected as the optimum.

Table-4: Investigation of optimum concentration of fish stock.

Stock solution Molarity (M)	Milli Volt before charging	Milli Volt after charging
0.1	241	278
0.2	240	260
0.3	220	260
0.4	200	230
0.45	185	215

Table-5: Investigation of optimum Concentration of Oxytocin.

Concentration of oxytocin (ppm)	Milli Volt before charging	Milli Volt after charging
0.01	99	210
0.02	105	190
0.03	100	132
0.04	80	121
0.05	90	180
0.06	113	130
0.07	114	224
0.08	190	264
0.09	240	285
0.10	277	305
0.15	287	310
0.18	258	310

Investigation of the power of fish scales battery

The stability and power of the phenol battery in our earlier work was investigated using a five cell battery [4] therefore, in the present study, a five cell battery was used for the investigation of the power of fish scale battery. The load for this battery was LEDS of the specifications given as: 1 V = 800-1000, V F (V) = 3.2-3.8, power dissipation = 80 mW. This battery was prepared under the conditions investigated earlier except the charging was carried out using a 36 volt charger. The results are given in Table-7.

Table-6: Investigation of the power of urea protein battery.

S. No.	No of LEDs	Power dissipation time
1	01	15 minutes and 10 seconds
2	02	12 minutes and 30 seconds
3	03	10 minutes and 55 sec
4	04	8 minutes and 10 sec

Conclusion

The fish scale battery was found to give a stable voltage. It uses a very small quantity of the oxytocin. Its voltage may be improved by proper measures like electrode material and the viscosity of the solution. The voltage of this battery is a function of the strength of the power supply for charging, the concentration of the peptides and fish scale solution. It is expected that this battery can be made into a useful rechargeable battery by the use of proper inert coagulant which increases the viscosity of the medium. It can also be used as a base for the electrochemical measurement of the antioxidant activity.

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