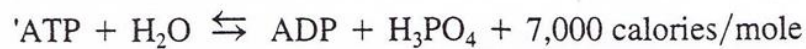


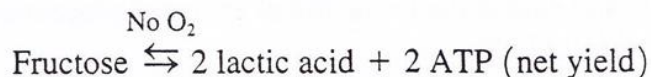
ENERGY METABOLISM BY SPERMATOZOA

Energy metabolism is the means by which spermatozoa convert energy substrates into usable forms of energy. Enzymes for this conversion are in the mitochondrial sheath. In addition to fructose, sorbitol, and GPC, which are present in seminal plasma, plasmalogen, a lipid found within the spermatozoon is an energy reserve that can be used when other substrates are limiting.

Adenosine triphosphate (ATP), a high-energy compound, is the form of energy that can be used by spermatozoa. It is converted to ADP yielding 7,000 calories per mole of energy by the following reaction:

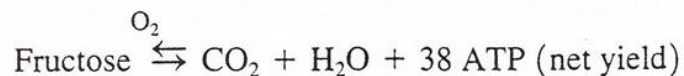


If there were no means of regenerating ATP, the spermatozoa would not survive due to lack of energy. Energy substrates provide a means by which ATP can be regenerated from ADP plus inorganic phosphorus. Fructose serves as a good example, since it can be utilized anaerobically and aerobically. The anaerobic reaction is as follows:



Fructose metabolized anaerobically yields a net of 2 ATP, or 14,000 calories. This reaction provides energy to maintain the viability of spermatozoa during storage. However, an end product of this metabolism is lactic acid. If steps are not taken to slow metabolism during storage, the buildup of lactic acid will soon lower the pH of the semen, adversely affecting the viability of spermatozoa.

Under aerobic conditions, the metabolism of fructose is



When oxygen is present, metabolism of fructose is 19 times more efficient in terms of energy yielded. The net energy from 38 ATP is 266,000 calories. When sufficient oxygen is present, the fructose molecule is metabolized completely to carbon dioxide and water. There is no buildup of lactic acid. In addition, sorbitol, plasmalogen, and, if in the female tract, GPC are available for metabolism and regeneration of ATP. Sorbitol and GPC are metabolized through the same biochemical pathways as fructose. Plasmalogen, a lipid rather than a carbohydrate, utilizes different metabolic pathways, but the needed enzymes are in the mitochondrial sheath.

14-4 FACTORS AFFECTING RATE OF METABOLISM

Rate of metabolism is the rate at which spermatozoa utilize their energy substrates. Under aerobic conditions, it can be monitored by measuring oxygen consumption, by measuring liberated carbon dioxide, or by methylene blue reduction. Under anaerobic conditions, the rate of reduction of pH or chemical determination of lactic acid buildup and/or fructose disappearance can be used as measures of metabolic rate. Control of metabolic rate is of interest because a reduction in metabolic rate is necessary to extend the storage life of semen. A number of factors contribute to reduced metabolic rate and extended life of spermatozoa in the epididymides (Chapter 3). In the epididymides, spermatozoa may remain fertile for up to 60 days. However, spermatozoa in a fresh ejaculate of semen will be fertile only for a few hours if steps are not taken to reduce their metabolic rate. The measures used must be reversible without injury to spermatozoa if they are to be practical for semen handling.

14-4.1 Temperature

Metabolic rate increases and the life span of spermatozoa decreases as the temperature of the semen rises. When the temperature rises above 50°C, spermatozoa suffer an irreversible loss of motility. If maintained at body temperature, spermatozoa will live for only a few hours due to exhaustion of available energy substrates, drop in pH due to buildup of lactic acid, or a combination of these factors. Reducing the temperature of the semen will slow metabolic rate and extend the fertile life of semen if precautions are taken to protect against *cold shock* and *freeze kill*.

14-4.2 pH

A pH of about 7.0 (6.9 to 7.5 for different species) falls in the optimum activity range of most of the enzymes in spermatozoa. Therefore, a higher metabolic rate is expected when the pH of semen is maintained near neutrality (7.0). If the pH of semen deviates toward alkalinity or acidity, metabolic rate will be reduced. The practicability of altering the pH of semen to extend its life is limited by the narrow range over which pH can be altered without permanently reducing activity. Research in this area has established the importance of diluting semen in a buffered medium that resists changes in pH, so that maximum fertile life of the semen can be maintained.

14-4.3 Osmotic Pressure

Semen maintains maximum metabolic activity when diluted with an isotonic diluter. Either hypotonic or hypertonic diluters will reduce metabolic rate, but neither will extend the life of the semen. The spermatozoon membrane is a semipermeable membrane. Both hypotonic and hypertonic diluters will alter transfer of water through this membrane, disrupting the integrity of the cell. It is very important that only isotonic diluters be used. Spermatozoa remain motile longest when suspended in isotonic media.

14-4.4 Concentration of Spermatozoa

Increasing the concentration of spermatozoa above that found in the normal ejaculate will decrease metabolic rate. Potassium is the principal cation in the sperm cell, whereas sodium is the major cation in seminal plasma. Increasing the cellular concentration will increase the potassium-to-sodium ratio in the semen. Potassium is a natural metabolic inhibitor. Increasing its concentration will reduce the metabolic activity in the semen.

Generally, moderate dilution of semen in a buffered, isotonic medium containing fructose will not greatly alter metabolic rate but will extend the life of the semen. Dilution such as this is usually done before lowering the temperature of semen. Some caution must be observed. If dilution is excessive (> 1 to 1,000), motility and metabolic rate will be depressed.

14-4.5 Hormones

Testosterone and other androgens depress metabolic rate, but those concentrations found in the male system have no permanent effect. Fluids from the female tract increase the metabolic activity of spermatozoa. This is thought to be primarily an effect from estrogens, but other unidentified factors may be involved. The increased metabolic activity in the female tract likely increases motility, which increases the frequency of collisions between spermatozoa and the oocyte in the oviduct.

14-4.6 Gases

Low concentrations of carbon dioxide stimulate aerobic metabolism of spermatozoa. If the partial pressure of carbon dioxide exceeds 5% to 10%, metabolic rate is depressed. Carbon dioxide has been identified as a factor in regulating metabolic rate in the epididymides. Oxygen is necessary for aerobic metabolism. On the other hand, too high a level of oxygen is toxic and will depress metabolic rate. This is not likely to be a factor in the laboratory unless oxygen or air is being bubbled through the semen. Anaerobic metabolism can proceed under nitrogen, hydrogen, or helium gases with no effect on metabolic rate.

14-4.7 Light

Light intensities that are normally found in the laboratory can depress metabolic rate, motility, and fertility in spermatozoa. The harmful effect is observed only if semen is in contact with oxygen. The enzyme catalase will prevent the harmful effect of light, which suggests that light causes a photochemical reaction in the semen that results in the production of hydrogen peroxide. Semen should be protected from light and never exposed to direct sunlight.

14-4.8 Antibacterial Agents

Gentamicin, Tylosin, and Linco-Spectin (Section 16-3) are added to semen during processing to control bacterial growth. None have a demonstrated effect on metabolic rate. They sometimes increase fertility of semen from low fertility bulls. Also, these antibacterial agents may extend the fertile life of the semen by controlling bacteria, thus sparing energy substrates for spermatozoa.