The choice and use of polymers in animal cages
By Leon Ernst.

The cost of plastic cages is one of the significant expenses in rodent facilities both as an initial purchase and as an ongoing replacement expense.
My intention is to assist you in your choice of polymer and then what you can do to reduce cage failure and extend cage life.

Choice of polymer.

Polymers have two family groups:

- **Crystalline polymers**
- **Amorphous polymers**

Crystalline polymers have good chemical resistance but generally can't be transparent (e.g. Nylon, Polyethylene (PE), Polypropylene (PP), Acetal)
Note there are transparent grades of clarified PP that are heavily nucleated to give small nuclei

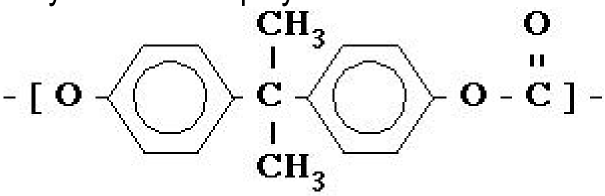
Amorphous polymers have poor chemical resistance due to their 'open' structure (that is high free volume) and are generally transparent (e.g. Polystyrene, Acrylic, Polycarbonate, Poly sulphone, PVC)

The only crystalline material in significant usage in cages over the years has been polypropylene but it has:

- **Poor clarity (can be made clearer by nucleation)**
- **Limited temperature tolerance**
- **High distortion probability**

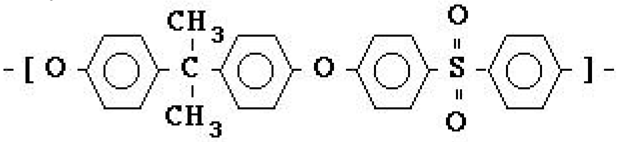
All of the now commonly used polymers are amorphous namely:

Polycarbonate and
Polycarbonate copolymer

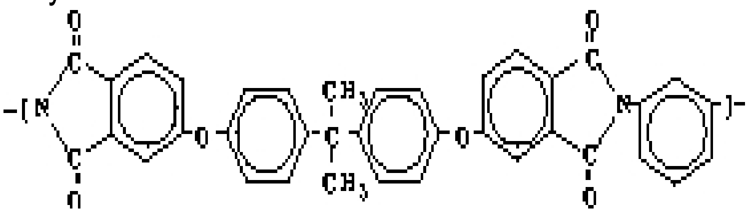


Note Polycarbonate co-polymer has a heat tolerance to 180 degrees C compared with standard polycarbonate at 120 degrees C

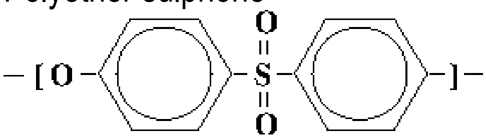
Poly sulphone



Polyether imide



Polyether sulphone



The cost of these materials in today's market is typically

Polypropylene	$2.20 per Kg
Polycarbonate	$6.00 per Kg
Polycarbonate co-polymer	$16.00 per Kg
Polysulphone	$25.00 per Kg
Polyether sulphone	$28.00 per Kg
Polyether imide	$38.00 per KG

With a typical cage weighing 500 to 600 grams it can be seen that the material content cost ranges from $1.50 to $23.00 per cage.

The cost to mould in each material does not differ significantly because this is simply the cost of cycling a machine.
To invest in the really expensive materials must yield a lifetime – cost benefit otherwise there is little point.

The use of the polymer (The causes of failure and how to minimise failure)

Why do these materials fail?

Firstly
All of these materials are at high risk of damage from a wide range of chemicals that can cause Environmental Stress Cracks (ESC). ESC occurs when opportunistic solvent molecules permeate the free volume of amorphous polymers and the swelling dilational stresses cause micro cracks to propagate.

Simply put, ESC is a failure mechanism whereby a plastic resin is affected by a chemical agent while simultaneously under the influence of tensile loading. It is a solvent-induced failure mode brought about by the synergistic effects of the chemical agent and the mechanical stress. The rate of generation of ESC is temperature dependant

Secondly
All of these materials are at high risk of damage from Ammonia (amines) and alkalis that can cause molecular chain breakage at room and elevated temperatures. The end result is similar to ESC but is not connected with stress.

ESC

You will see evidence of Environmental Stress Cracking in polycarbonate Kitchenware characterised by a myriad of extremely fine cracks (ie. "silver cracking") that initially are very shallow and, in the early stages only appear in some lighting conditions. Detergents used in the kitchen sink or in dishwashers almost always cause this phenomenon. Once these chemical stress cracks start, failure follows rapidly.

A clear distinction can be drawn between the **inherent stress** in the cage mouldings and the **induced stress** on the cage mouldings.
By way of definition: **inherent stress** is the ‘in-built stress’, 'frozen-in stress' or ‘residual moulded-in stress’ formed during the manufacturing process; while, **induced stress** is the externally applied stress that arises from handling, storage and stacking and general use after the product is manufactured.

Susceptibility of Cage Polymers to ESC (Environmental Stress Cracking)

All of the cage polymers are amorphous, meaning they have a high level of free space in their structure allowing ingress of opportunistic molecules such as greases and oils. Environmental Stress Cracking is a prime cause of failure for polymer cage products.

The most likely chemicals that will cause ESC in cages are in detergents and surfactants and include **vegetable oils like pine oil or lemon oil (d-limonene)** and **glycol-ethers** used to cut through grease. These chemicals on their own or in combination will cause ESC

Stress plays a vital role in Environmental Stress Cracking. In simplistic terms

E x S = C

Where
E is the environmental agent (chemical),
S is the stress and
C is the cracking.

Without the stress in the equation no cracking occurs. Significantly the stress required for ESC to occur is much lower than the yield stress or break stress of the polymer.

Fundamentally, **ESC** results from the combined action of an environmental agent and stress where the stress can be either an in-built (inherent) stress or externally applied (induced) stress that exceeds a critical threshold (for example, 500KPa). Thus in the absence of **E** (or under the application of a very mild **E**), Environmental Stress Cracking will not eventuate; likewise when **S** is below the threshold there will be no cracking. It is mandatory for both **E** and **S** to be present in order for there to be Environmental Stress Cracking

Since caging materials are well known in the industry to have high susceptibility to ESC problems, best practice with regard to the plastic moulding manufacturing operation is vital to ensure that this form of failure is minimised. In particular steps should be taken to minimise built-in stresses by optimisation of parameters such as cycle time, melt temperature, mould temperature and more importantly by performing proper annealing operations post-moulding.

It is important to note that with ESC there is no chemical attack or molecular degradation. Conversely, the chemical permeates into the molecular structure and interferes with the intermolecular forces binding the polymer chains, causing molecular disentanglement. ESC is a brittle fracture mode and the steps involved in the mechanism include:

- **Fluid absorption,**
- **Plasticisation,**
- **Craze initiation,**
- **Crack growth, and finally**
- **Fracture.**

In terms of growth mechanism, it is said that gaps open between molecules when the part is loaded or in some other condition of stress, that the chemical agent penetrates into these gaps, and that the cohesive power (or strong binding) between the molecules is reduced.

Alkalis and Ammonia and Direct Chemical Degradation)

Ammonia (amines) and its affect

Urine rapidly decomposes to produce ammonia and some mouse disorders also produce urine with high initial ammonia content

Ammonia is a cause of cage failure and is different from ESC but produces the same outcome.
Ammonia and amines (particularly secondary amines) cause degradation of cage polymers via aminolysis. That is cleavage of polymer chains by ammonia. Chain cleavage reduces molecular weight and therefore polymer strength. Eventually the polymer if impacted, will shatter like glass.

It is usual practice that all detritus is removed from a cage before autoclaving. It is the method of removal that will influence the life of the cage.

Many institutions have found that hot water only washing is able to achieve a high standard of detritus removal and this is by far the best process.

The down side is that simple water washing may leave behind trace urine residues that at autoclave temperatures will be damaging to the cage materials.

It is because of these residues that detergents or surfactants are often used

The benefit of detergents is offset by the increased risk of ESC and damage is compounded by high concentrations of detergents on the belief that "more detergent gets it cleaner".

Alkalis and their affect

All of the cage materials are degraded by alkalis. Polysulphone and Polyether imide are particularly susceptible to alkaline hydrolysis. The degradation is independent of intrinsic or applied stress but is accelerated dramatically by temperature.

What you can control is the chemistry of cleaning

Dishwasher detergents are generally highly alkaline Sodium metasilicate based and often have vegetable oils in them like pine oil or lemon oil (d-limonene) or some other material to provide protection for skin. This is probably an urban myth with regard to protecting skin but it sells detergents and it can provide additional surfactant properties in the cleaning process. Also detergents contain glycol-ethers to cut through grease.
These chemicals have a combination of materials that will cause ESC and Chemical degradation because these vegetable oils, Glycol-ethers, and alkalis will all cause severe damage to all of the cage polymers particularly at autoclave temperature.

While these chemicals can seriously damage cages, the most significant risk comes from the cage contents.

Cage contents at change over are bedding, urine residue and faecal matter often in a semidry state.

What should be done to achieve maximum cage life

1. Start the wash process as soon as possible after change-over
2. All detritus must be mechanically removed from cages
3. Cages must be washed in hot water (70 to 80 Deg C) with mechanical scouring
4. Cages must be rinsed in clean water with a pH less than 7
5. Cages must be dry and free from stains prior to autoclaving

Sometimes complete removal of detritus is not achieved and this is justified on a cost basis or can be part of a protocol whereby bedding is deliberately autoclaved in the box. If you must do this, the boxes will be destroyed in a few cycles of autoclaving

Most of you will have noticed that cage bases degrade far more rapidly than cage lids. This is simply because lids never come in direct contact with urine (ammonia).

It must be said that no matter how good the cage cleaning, the bases will degrade before the lids because urine (Ammonia) at room temperature will still initiate degradation and lead to cracking and failure.
The majority of chemical reactions are concentration - time – temperature continuums and a cage at room temperature for 7 days with urine/ammonia contact will start to degrade as previously described. You can’t do much about this but the same factors in one autoclave cycle at temperatures of 121 or 134 can be equivalent to several weeks at room temperature.

There is some evidence that the choice of bedding can reduce cage failure. There is need for a good study here but anecdotal information suggests that reconstituted paper is likely to increase failure rate while corncob seems to decrease failure rate. This could be associated with the labile pH of the bedding in that paper is alkaline while corn cob is neutral or slightly acidic. My observation from this is that the neutralising affect of low pH bedding may be beneficial. I am inclined to wonder what possibility there is to produce bedding with a pH of about 2 or 3 to aid in neutralising the ammonia from the urine. The down side is that the act of neutralising may well produce new tertiary amines

Mechanical scouring can be achieved by vigorous jets of hot water in a cage washing machine or by old fashioned scrubbing with a non abrasive sponge.

The final rinse is critical and in today’s limited water supply and conservation environment, rinsing is often less than satisfactory with rinse water being little different from wash water. Again I suggest that a low pH rinse may be beneficial.

A dry cage will clearly show if there are residues.

This brings us to autoclaving.

It can be shown, and you can easily do this in your facility, that autoclaving alone does not destroy cages.

Simply get a new cage and recycle it through the autoclave every time you use the autoclave without the cage going into service in a mouse rack. This will demonstrate that even after several hundred cycles and more there will be little sign of damage with any of the materials commonly used. There may be small changes in colour and some materials may distort slightly but otherwise the cages will be fit for their designed application.

This brings us back to cleaning and associated cost.

If through poor cleaning the cage life is halved then this can be seriously expensive.

The process of cleaning is in itself expensive. You will all have a clear view of your cost in your operation and you have probably spent a lot of time minimising this cost.

The added cost of cage replacement is where you can with little expenditure make large financial savings.

Research funding is not always easy to get. You all have the day-to-day running expense of food, water, operations, staff, services etc. sometimes the longer term savings appear ephemeral. The savings that can be achieved with cages amortised over four years instead of two is simple arithmetic. Do your sums and then see how little you need to spend to get a real financial benefit.

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