

 **APL** BIOPURIFICATION
TECHNOLOGIES

Corporations can benefit from a better way to purify biomolecules. Reducing the time to evaluate and develop new purification processes, and lowering development and production costs are often the critical success factors in **developing blockbuster compounds**.

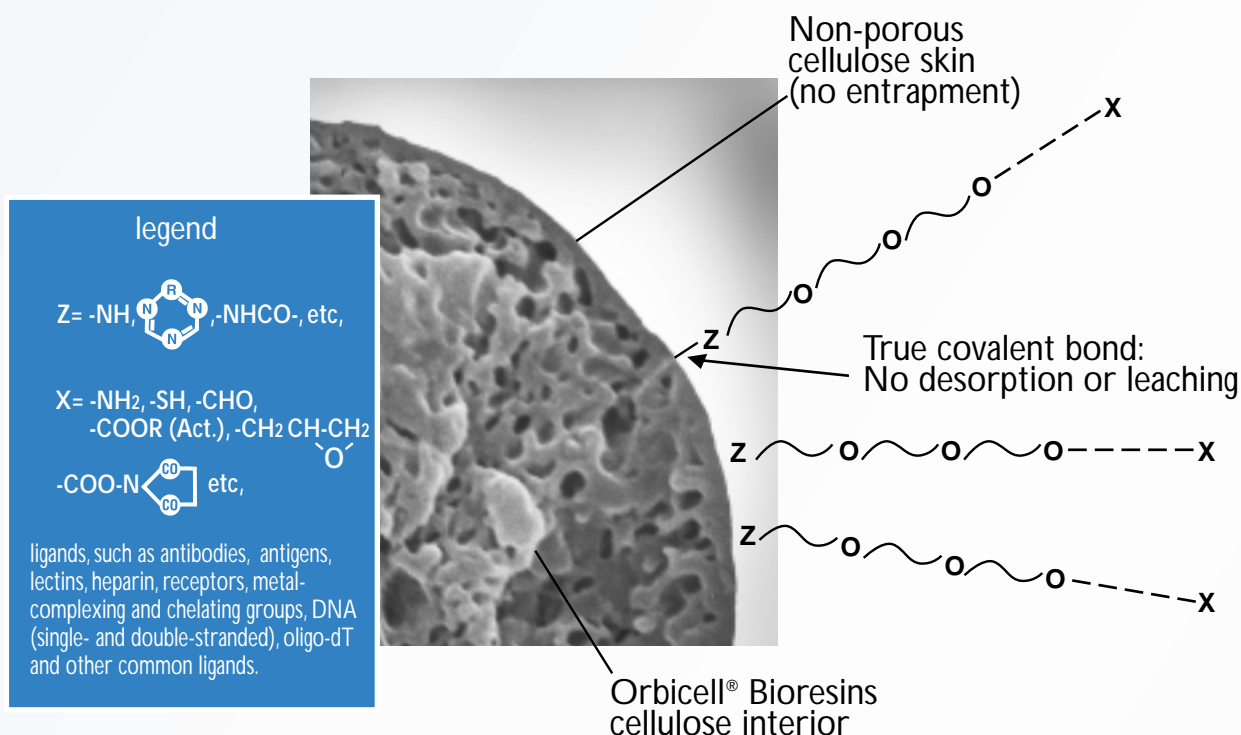
APL Biopurification Technologies provide a unique and patented glassy, non-porous matrix that allows quick separation of large quantities of target molecules from complex mixtures resulting in high yield, high purity products.

Revolutionary Orbicell® Bioresins and Continuous Flow Purification Processes

APL Biopurification Technologies (APL's) matrix, Orbicell® is a matrix of spherical, glassy non-porous beads that withstand over 700 psi pressures and wide pH ranges. When the physical properties of this robust matrix are combined with APL's unique chemical binding properties and proprietary continuous centrifugation and cross flow purification processes, proteins can be purified more efficiently than with traditional methods. A wide range of ligands has been attached to the Orbicell® matrix for both purification of target proteins and other biomolecules. The proprietary surface chemistry covalently bonded to the Orbicell® bioresin enables very high capacity for proteins (in some cases reaching up to 10% of the dry weight of the bioresin), eliminating leaching which arises from less stable attachments which can occur with currently available separation matrices.

Excellent yields of very high purity products can be isolated because of the way the Orbicell® bioresin is constructed. The extremely low non-specific interactions of the novel glassy cellulose surface may account for the unusually high (85 to 95%) first pass purities. Its robust physical properties allow for use in high-throughput, continuous flow applications such as centrifuge or cross flow systems.

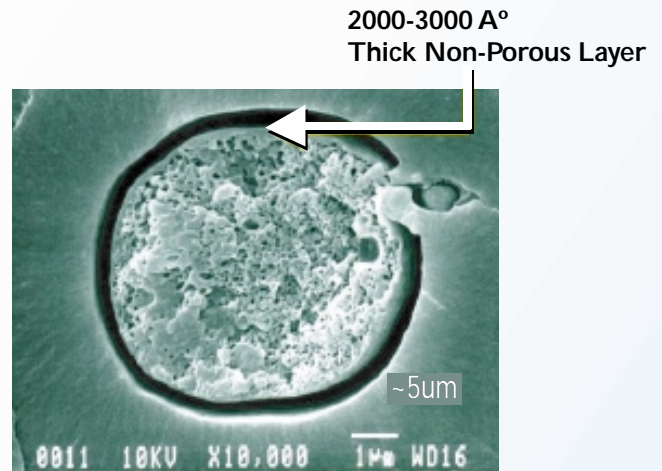
Orbicell® Unique Cellulose Affinity Bioresins



Electron Micrograph of Orbicell® Bead

Molecules that are over 1kDa do not penetrate the bead, thus eliminating band broadening due to molecular sieving effects.

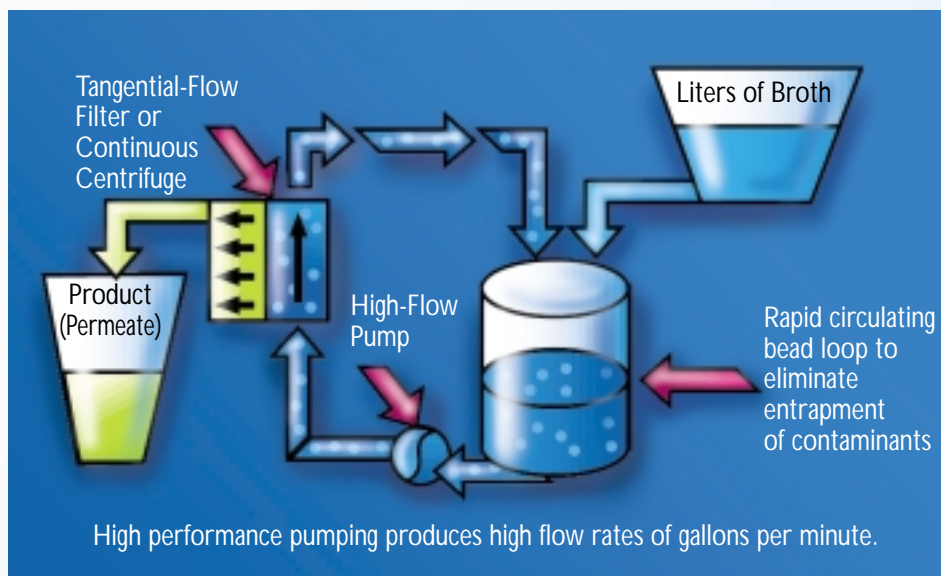
The process of washing and releasing target proteins bound to a solid, non-porous matrix in a cross flow system allows **better separation of target molecules and removal of contaminants.**



Benefits of Orbicell® Affinity Bioresins

- Small organic affinity ligands are attached to the cellulose support through **stable covalent bonds eliminating leaching problems**
- Affinity ligands are **not susceptible to proteolytic cleavage**
- **Loading** under saturation conditions ensures high capacity, up to 10% of the antibody weight per dry weight of the sorbent
- The **non-porous character** of the Orbicell® bioresin **eliminates entrapment** of non-target molecules allowing for quicker washing and production of purer end products
- Orbicell® bioresins routinely **achieve purifications of 85-95% first pass**
- Orbicell® bioresins can be **sterilized by autoclaving** (no other chromatography support we've seen can do this)

Multikilogram Recovery of Biomolecules using Orbicell® Affinity Bioresins



Contaminants Pass Through While Orbicell® Bioresins Retain Target Molecules

Withstands High Flow Rates of Tangential Flow Systems or High Pressures of Continuous Centrifuge Processes.

Orbicell® Affinity Bioresins and Applications

Orbicell® Resins	Application
Protein A	Purification of monoclonal or polyclonal antibodies
Simul-A or Simul-M	Purification of monoclonal or polyclonal antibodies without using Protein A (susceptible to enzymolysis) as an affinity hook
Metallo bioresins	Purification of poly-histidine fusion proteins and antibodies
Streptavidin	Purification of biotinylated nucleotides and proteins
Reactive Blue Dye	Purification of human serum albumin, coagulation factors, interferons, and other biomaterials
Benzamidine	Purification of activators, urokinases and serine proteases
Activated Ester	Covalently attaches binding materials such as cancer antigens, antibodies, viruses allowing in-situ custom derivatization without wet chemistry
Hydrazide	Common reactive group for easy ligand attachment to beads
Aldehyde	Common reactive group for easy ligand attachment to beads

Physical Characteristics and Benefits of Orbicell® Affinity Resins

Characteristics	Benefit
Stable in organic solvents, even when starting in water and compatible with 8M urea, and chaotropic reagents like 6M guanidine-HCL	Increases options for ligand attachment; also complex solid phase reactions in anhydrous organic solvents are now feasible Wide options for binding and elution of target molecule
Does not shrink in pure organic solvents	Fully accessible outer surface for attachment of ligands
Rigid bead structure	ROBUST - Can be used under demanding conditions Tolerates continuous medium pressures up to 700 psi and intermittent high pressures Physical integrity not compromised under rigors of high speed centrifugation or cross-flow filtration allowing many cycles of reuse
Solid, non-porous, bead with glassy surface	Rapid removal of unwanted material, no entrapment in interior of beads Simpler, much quicker, purification scheme
Operates in wide aqueous pH (1-13) conditions	Efficient and more complete cleaning between each use including sterilization by autoclaving with no significant degradation of the beads
Minimal expansion in volume in water (less than or equal to 15%)	Rigid structure maintained; no pores to be clogged