

## Challenges and Opportunities of Monoclonal Antibody Manufacturing in China

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### Introduction

Monoclonal antibody (mab) drug represents a major class of biotechnology therapeutics. Currently there are 22 FDA approved mabs with global sales reaching \$26 billion in 2007. Data monitor predicts that mab sales will continue to grow exponentially and reach \$50 billion in 2013 while mab sales in 2000 was barely \$2 billion. Among the FDA approved mab class drugs, including full mabs, Fc fusion proteins, pegylated Fabs, all but one of these proteins are produced in mammalian cell expression systems using Chinese hamster ovary (CHO) or murine hybridomas (e.g., NS0, Sp2/0). It is estimated that about 200 mab candidates are in clinical development, and that these candidates will fuel future growth of this important drug class <sup>[6]</sup>.

### Success of monoclonal antibody therapeutics in the US and EU

Antibody drugs have achieved phenomenal success in oncology with Rituxan, Herceptin, Avastin, and Erbitux all achieving megablockbuster status (several billion dollars of sales) and in autoimmune diseases where Enbrel, Remicade, and Humira have combined sales of more than \$16 billion in 2008. Clinical and commercial success of monoclonal antibodies has mandated a need for large-scale production in mammalian cell culture, which has resulted in rapid expansion of global manufacturing capacity and an increase in size of bioreactors up to 25,000 L. Global protein output using mammalian cell culture in 2000 was less than 500 Kg while in 2005 the number jumped to 3600 Kg. It is estimated that in 2007 the annual demand of bulk drugs of Enbrel, Remicade and Rituxan all exceeded 700 Kg, respectively, which is significantly higher than each of the products' global output of less than 500 Kg in 2000. In addition, compared to the total output of 3600 Kg of mabs, the total output of non-mab glycosylated proteins (such as EPO) is only approximately 60 Kg.

As mabs become hugely successful in the clinic, a greatly increased effort to improve process efficiency with concomitant manufacturing cost reduction was implemented. This has been particularly successful in the cell culture process where productivity has improved 100+ fold in the last 15 years. It is estimated that mab titer almost doubles every 18 months, an exponential growth rate often referenced in the electronics industry as the "Moore's Law". This success is resulted from the improvements in 3 major areas: expression technology, media optimization, and advanced fed-batch culture technology. In addition to improving process/cost efficiencies, platform technologies are being developed to reduce the time to develop processes and produce the first batch material required for clinical testing. As all biotech and large Pharma have endorsed mabs as their key future directions, competition in the development of mab therapeutics is becoming more intense. It is estimated that 10 years ago the second player of a follower mab with the same target of the first player usually lags by 3-5

years. Recently it is reported the lag has been reduced to 12-18 months. Furthermore, a month delay of going into an clinical trial results in a delay of at least a month of launching the drug, which means a lost revenue of \$83 million for a blockbuster drug, not to mention the possibility of an earlier launch by competitors. Therefore mab drug development has become a marathon race with multiple 100-meter sprints. One needs to win both the marathon and the sprints to be highly successful.

### Challenges of mab manufacturing

Unlike traditional small molecule therapeutics that are produced through chemical synthesis for less than \$1 per gram, mabs must be produced in mammalian cells at a cost of \$300-\$5,000 per gram. Although simple proteins/peptides such as insulin, growth hormone, and G-CSF are produced in bacterial hosts such as *E. coli* with various advantages such as established regulatory track record, well characterized product and competitive cost; complicated proteins with glycosylation mostly can not be produced in *E. coli* because *E. coli* is unable to modify proteins after they are produced and it often makes misfolded and, thus, inactive proteins. For these reasons, monoclonal antibodies and fusion proteins can not be produced in *E. coli* but rather must be produced in mammalian cells. Compared to *E. coli* system, the construction of the genetically engineered mammalian cells is more time-consuming, production is less efficient/more expensive, and product launch requires an expensive commercial scale manufacturing capacity.

Mab development begins with the construction of a master cell bank. This involves genetically engineering a host mammalian cell, typically a Chinese hamster ovary (CHO) cell, to produce the protein of interest. CHO cells are often used because they multiply quickly, are relatively hardy, and grow well in suspension culture. When a cell line is created that produces high levels of the desired protein and can be grown robustly in suspension cell culture, a master cell bank is created. It will serve as the source of all cells used in the production of clinical and commercial quantities of protein under development. The creation and characterization of the master cell bank typically takes 6-8 months.

Once a master cell line has been created, the next step is bioprocess development. This covers 2 phases of activity: expansion of the cells to a large volume and recovery and purification of the target protein. Currently there are two methods used to grow cells: fed-batch mode and perfusion mode with more than 90% of processes using fed-batch culture. In fed-batch runs, cells derived from the master cell bank are progressively grown in larger and larger volumes over a period of 3 to 4 weeks to provide a

seed culture for large production bioreactors. This gradual step-up in volume allows for the most rapid growth of a large volume of cells. The production bioreactor typically holds between 10,000-20,000 liters of culture medium. Once the seed culture is added to the production bioreactor, the cells are grown to an optimal density over a period of 10-20 days. At this point, the cells are collected by filtration and the protein of interest is then purified through 3 chromatography steps. In addition, processes to inactivate and remove viral contaminants must also be included in the production process to ensure patient safety. The final, purified product is then formulated and filled. The purification process takes about 5-10 days on average while the entire manufacturing process takes approximately 8-10 weeks per batch. Typically CHO cells produce 1 gram of product per liter of cell culture medium. For monoclonal antibodies, a typical purified bulk yield is ca. 60%. This translates into 6kg of finished product per 10,000 L bioreactor. Currently the largest mammalian cell culture manufacturing facility is Genentech's Vacaville facility with 200,000 L of capacity.

Once the clinical development is complete and the drug is approved, managing appropriate manufacturing capacity is extremely difficult. Too little capacity leads to lost sales while too much capacity results in unwarranted capital expense, low utilization rate of the facility, and high depreciation cost. Mab manufacturing took center stage in the late 1990s as a stream of monoclonal antibodies (e.g., Genentech's Rituxan and Herceptin, and MedImmune's Synagis) and antibody-fusion drugs (e.g. Amgen/Immune's Enbrel) achieved unprecedented success in the market. While this unforeseen success was welcome, the widely known Enbrel shortage as well as other manufacturing-related product delays highlighted the importance of adequate planning for manufacturing capacity<sup>[8]</sup>. Enbrel's shortage mandates that patients already on Enbrel had to wait in line to get their drugs and no new prescriptions were allowed for new patients. It is estimated that Immunex potentially lost billions dollar of Enbrel sales and also allowed faster uptake of a competitor product Remicade from Johnson & Johnson during launch.

### Mabs in China

Despite that development of antibody drugs is in full swing in the US and EU, new antibody drug development in China is difficult. A review of launched mabs in China shows that most of the mabs achieving >100 million RMB sales (Chinese standard of blockbuster drugs) are from multinationals except for biosimilar etanercept. So far all the mab drugs from multinational companies such as Roche's rituximab, trastuzumab, and Novartis's basiliximab have achieved blockbuster status

in China with as high as 50% annual growth rate except a recently launched product Remicade. Meanwhile, the only blockbuster mab drug made by Chinese company is biosimilar etanercept, which brought in approx 140 million RMB sales in 2008, 3 years after its launch in China (IMS Health, 2009)

A survey in China shows that most Chinese players are still focusing on proteins expressed in microbial systems. It is reported that there are 500 protein drugs in clinical development in the US and EU, out of which approximately 70% is glycosylated proteins that are expressed in mammalian cell culture systems<sup>[2]</sup>. However, a majority of the protein therapeutics in the clinics in China are expressed in microbial systems<sup>[4]</sup>. In the past couple of years, there are rapidly growing interests in mab de-

velopment. However, high technical barriers of entry prevented Chinese players from leapfrogging to the most advanced stage of mab development. Fundamental reasons for this slow progress are lack of novel antibody products, lack of efficient cell line construction technology, and difficulty in large scale antibody production (i.e. large-scale, high-density, high expression mammalian cell culture technology).

#### **Lack of novel products**

Mab development in China started mainly in academic research institutions in the late 1980s. However, there is little progress since then. Currently fewer than 10 mabs in the various stages of clinical trials are developed by Chinese domestic players. While all of the mabs enter-

**Table 1:** *launched mab products in China*

Generic name	Trade name	Type of molecule	Sales (> 100 million RMB)	Sponsor
Muromonab	OKT3	Murine	Yes	Johnson & Johnson
Rituximab	Rituxan	Chimeric	Yes	Roche
Trastuzumab	Herceptin	Humanized	Yes	Roche
Basiliximab		Chimeric	Yes	Novartis
Cetuximab	Erbitux	Chimeric	Yes	E Merck
Infliximab	Remicade	Chimeric	No	Johnson & Johnson
Mouse Anti-human CD3		Mouse	No	Wuhan Institute of Biological products
Anti-human IL-8 mAb cream		Murine	No	Dongguan Hongyuan
[131I] chimeric tumor cellular nucleus mAb (131I-chTNT)		Murine	No	Shanghai Meien Biotechnology Co.
Iodine (131I) Metuximab (131I-Anti CD147)	LICARTIN	Chimeric	No	Sichuan Huashen Co.
Nimotuzamab (Anti EGFR)	Taixinsheng	Chimeric	No	Beijing Biotechnology Co.
Etanercept (TNFR-Fc)	Yisaipu	Fc Fusion protein	Yes	Shanghai CP Guojian

ing clinical trials in the US and EU are humanized or fully human, most of the programs in China are still chimeric. Several of them are still mouse mabs. Lack of novel products and humanization technology are strong hindrance of the development in this area. Therefore Chinese players are mainly interested in biosimilar mab programs.

### **Low expression level of engineered antibodies**

Except a few antibody fragments such as Fabs that can be expressed in yeast or *E. coli*, most antibodies require mammalian cell culture to ensure proper post-translational modification and subsequent bioactivity. Many laboratories in China can only achieve antibody expression in 10 mg/L range, which makes production cost-prohibitive. A few players have technologies that support antibody expression in 100 mg/L range with few achieving more than 1 g/L. In the US and EU, the expression levels of antibodies are typically higher than 1 g/L in commercial production and reaching as high as 10 g/L level in labs.

The main reasons for the low mab expression in China result from 2 factors: lack of cell line construction expertise and lack of experience in cell culture process development (media/feed and process). Cell line construction and selection is often a critical path and needs to be completed rapidly without compromising quality. Productivity of mammalian cell culture in the US and EU has improved dramatically in recent years. Highly productive cell lines result from using a host cell line that has the desired characteristics, an appropriate expression system, and a good transfection and selection protocol. A number of expression systems with the potential to produce cell lines with high specific production rates ( $Q_p$ ) are available. The challenge is to create cell lines that not only have high  $Q_p$  but also can grow to high density. Ideally, one would like to rapidly create a highly productive cell line that could be used for long-term

manufacture obviating the need to create an improved second generation cell line at a later stage of development. In the cell culture development area, currently there is little research in China. Therefore, Chinese biotech companies are facing tremendous difficulty hiring experienced personnel to work on bioprocess development. In contrast, during the initial growth period of mab in the US in the late 1990s, an explosion of interdisciplinary research in biochemical engineering paved the way for the advance of cell culture development and also trained all the leaders currently working in this mab commercialization.

### **Large-scale mammalian cell culture technology**

Even if academic labs in China can create a cell line and develop a corresponding process that yields 1 g/L production, lack of expertise of process scale-up might still pose additional challenges for mab manufacturers in China. There is a huge gap in large-scale, high-density, high-expression cell culture technology in China. Due to technical and financial reasons, typical cell culture facility in China is at ~100 L bioreactor scale. Currently only 3 operating facilities have bioreactor capacities that exceed 1500 L, which is only considered as a pilot scale in the US and EU (Table 2). In addition, many of the Chinese players still employ microcarriers to grow adherent cells or use continuous perfusion mode to produce mabs while in the US and EU suspension culture of cells at fed-batch mode is preferred. Cell culture scale in the US and EU is normally ~ 15000 L. Considering the increase of cell culture productivity in recent years, 5000 L scale is currently sufficient for product launch and initial stage of commercial manufacturing.

Different from earlier mammalian cell culture products such as EPO and interferon, mabs typically require a much higher dose. Humira and Herceptin for use in autoimmune and oncology areas are typically used to estimate clinical requirements of bulk mab drugs. Dur-

**Table 2:** Chinese mab players with 1500+ L bioreactor capacity

	<b>Beijing Biotech</b>	<b>Shanghai CP Guojian</b>	<b>Shanghai Celgen</b>
Product	Anti-EGFR mab	Biosimilar etanercept	Biosimilar etanercept
Cell line	NS0	CHO	CHO
Cell culture	Perfusion	Fed-batch	Fed-batch
Bioreactor capacity	2x 750L	4x750L	4x500L
Annual capacity	5 Kg	20Kg	Undisclosed

ing their product development in the US, approx. 3.1 Kg and 3.2 Kg of bulk drugs were used in the clinic. It is interesting that a higher dose mab of Herceptin and a lower dose mab of Humira require almost the same amount of bulk drugs. The difference comes from more frequent dosing of longer treatment cycle of the lower dose mab of Humira. In addition, the need of bulk drug does not account for materials needed for development use including process development, viral clearance and stability studies, which can add 1-2 Kg of additional material requirement. Since the requirements for drug development in China are less stringent than in the US and EU and clinical trials in China typically require smaller patient population, approximate 0.8-1.2Kg of mab bulk drug is required to complete clinical development in China (Table 3). With average titer of 100 mg/L and average scale at 100 L, an unrealistic number of approximately 160-250 batches are needed to complete product development. The bottleneck of Chinese mab industry is in large-scale manufacturing.

Another piece of evidence manifesting this bottleneck is that Chinese biotech companies often delay product launch several years after they obtain approval<sup>[5]</sup>. The main reason is that these companies do not have the expertise in manufacturing complicated proteins such as monoclonal antibodies or fusion proteins. Delaying product launch for as many as 3 years can result in millions of lost revenue. Jia linked this to the fact that although China trains tens of thousands of life scientists each year, process development specialists are rare, which is part of the reason that the country is encountering a bioproduction bottleneck.

### Opportunities of mab manufacturing in China

Although there are tremendous challenges in growing a sophisticated mab industry in China, there are also enticing opportunities to serve the large domestic market, to provide contract manufacturing service to reduce cost of mab therapeutics in the US and EU, and to develop high-quality biosimilar mabs for the global market. Returnees from the west who can bring a wealth of product development experience from the US and EU can play a significant role in expediting mab development and manufacturing in China.

### Large unmet medical need in Chinese domestic market will fuel phenomenal growth

According to IMS Health's estimate, Chinese pharmaceutical market will rank 6<sup>th</sup> globally in 2011, changed from 9<sup>th</sup> in 2006. It is estimated that China will develop into a top 2 pharmaceutical market in 2020. Underlying the phenomenal market growth is sad statistics. The number of new cancer patients in China increases about 1.6 million while 1.3 million die annually. Breast cancer prevalence in urban cities has doubled over the past 10 years. There are more than 200 million hepatitis virus carriers in China, where 120 million are HBV carriers. The total number of diabetes patients in China is above 20 million with the incidence of diabetes increasing from 0.67% up to 3.21% in the past 20 years. To tackle these diseases, Chinese government is investing heavily in biotechnology. Since mab's growth in the US and EU is mainly due to its addressing unmet medical need, mab therapeutic is a new and promising industry with great potential and phenomenal growth in China.

**Table 3:** Example of clinical requirements for typical low/high dose mabs

	Phase I	Phase II	Pivotal Studies
Number of subjects for Humira®	200	600	2000
Clinical bulk of Humira®	0.2 kg	0.7 kg	2.2 kg
Number of Subjects of Herceptin®	50	700	500
Clinical bulk for Herceptin®	0.1 kg	1.8 kg	1.3 kg
Number of Subjects for autoimmune mabs in China	50	200	800
Clinical bulk for autoimmune mabs in China	0.05 kg	0.2 kg	0.9 kg
Number of Subjects for cancer mabs in China	50	200	400
Clinical bulk for cancer mabs in China	0.1 kg	0.2 kg	0.5 kg

### **High cost of mab treatment in US/EU will drive innovative companies to reduce cost**

The patient cost of mab therapeutics has become an increasing concern in the US and EU. Mab cost is high in general because of the high manufacturing cost and large investment needed to get the mab approved. A mammalian cell culture facility typically costs several hundred million dollars and takes 5 year to build. Raw materials used in mab manufacturing are expensive with the cost of Protein A affinity resin as high as \$17000 per Liter. In addition to high manufacturing cost, drug developers often must pay stacked royalties to those who hold key intellectual property in MAb production, humanization technology, and targets.

When Genentech launched Herceptin in 1998, the cost of typical treatment with the drug is ca. \$20,000 per year. The price attracted notice, but little criticism. Four years later, Bristol-Meyers-Squibb and ImClone Systems began charging as much as \$100,000 a year for Erbitux, a drug for advanced colon cancer. Later a year's supply of Avastin drug for an average colon cancer patient costs \$54,000. Because of the significant growth of health care cost in the US, these prices are unsustainable. Therefore innovative companies will need to find manufacturing sites with competitive cost structure, either in tax heavens such as Puerto Rico, Ireland or Singapore or in low-cost manufacturing countries such as China and India. Providing CMO service to large multinationals to reduce the cost of manufacturing mabs in the US and EU may be an intriguing business opportunity for China.

### **Chinese companies will have a significant play in biosimilars**

Most mab programs in the clinic in China are biosimilars. These programs will have opportunities outside of China if high-quality products similar to innovators' can be developed. There are currently approved pathways for biosimilars in EU, Canada and Japan. It is also anticipated that the US will approve biosimilar (also known as follow-on biologics) in the near future. Decision Resources, Inc., a leading research and advisory firm for pharmaceutical and healthcare issues, predicts that biosimilar TNF-alpha inhibitors for the treatment of autoimmune diseases will become the first \$1 billion biosimilar class across the US and Europe in 2014, so Chinese companies that have the technology know-how can play well in the biosimilar arena.

In addition to providing biosimilars for the US and EU market, substituting made-in-china mab products for imported drugs is an inevitable trend for Chinese patients.

Mab treatment is expensive in the US and EU, and it is even more so in China as most mab drugs are priced higher in China than in the US because of the limited market size and the additional expense of developing them in China. This inevitably prices 99.9% of Chinese patients out of the market as annual cost of the most affordable antibody drug is close to 50,000 RMB, which is significantly higher than the average annual income of the Chinese population. Therefore, made-in-China bio-similar mabs are in demand to address the unmet medical need in China. Only these drugs can fully realize the market potential of antibody therapeutics in China. The history of interferon and EPO development in China substantiates this trend. Since the launch of domestically produced interferon and EPO, the average price has come down to a tenth of its original cost.

### **Summary**

Development of monoclonal antibody therapeutics is in full swing in the US/EU. However in China it is a still nascent industry. Because of the huge potential domestic market in China and mabs addressing unmet medical need, mab therapeutic is a new and promising industry with great potential and phenomenal growth in China. Returnees with a wealth of relevant experience will play a significant role in help this nascent industry leapfrog to be able to compete with established companies in the US/EU.

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