White Paper

Derisking Bispecific Antibody Development with The B-Body® Platform

Abstract

Bispecific antibodies (BsAbs) offer therapeutic capabilities that are difficult to achieve with conventional monoclonal antibodies, but their structural complexity can introduce significant challenges during discovery, optimization, and manufacturing. The B-Body® bispecific platform—engineered by Invenra—was designed to reduce these risks by providing an IgG-like framework that promotes efficient heavy- and light-chain pairing while supporting the plug-and-play combination of diverse Fab domains and Fc modifications. Since Invenra's launch of this platform 8+ years ago, the data generated across thousands of B-Body constructs demonstrate consistent expression and purity across Fab pairings, compatibility with high-throughput screening and standard mAb manufacturing processes, and robust stability under a range of storage and handling conditions. B-Body molecules have also shown mAb-like pharmacokinetics, low immunogenic potential in comparative assays, and compatibility with high-concentration formulations. Together, these characteristics provide a path for evaluating bispecific candidates in a format that is designed from the outset with both functional screening and downstream manufacturability in mind, helping reduce the risk that promising molecules encounter avoidable development barriers later in the pipeline.

Introduction

Interest in bispecific antibodies (BsAbs) has grown considerably over recent years: As of July 2026, fourteen BsAbs have received approval for commercial use in the United States, twelve of which came within the last five years.[1] Their ability to simultaneously bind multiple epitopes enables BsAbs to do what few other therapeutics can, whether it's guiding immune cells to malignant tissues, inhibiting multiple signaling pathways, or delivering cytotoxic payloads with high precision. This versatility has led to an upswell of BsAbs in drug development pipelines, with potential applications ranging from cancer to cardiovascular disease and many others. Currently, there are more than 400 active clinical trials involving BsAbs, with many more expected in the coming years.[2]

However, developing a viable BsAb is a complex process that's prone to significant uncertainty and risk. Though there are many BsAb formats that researchers can choose from, all come with the same basic challenge of precisely engineering a stable protein complex, one that holds at least two distinct antigen binding domains. Such a process typically requires lengthy and costly optimization cycles.

Even when a well honed therapeutic candidate is found, logistical challenges may prevent it from advancing into clinical trials. It is not enough for the BsAb to have optimal pharmacological properties, it must also be ready for bulk manufacturing. The protein's design and manufacturing processes must often be further optimized to enable robust expression and purification, as well as enduring stability and many other properties. Put simply, a promising candidate means very little if it can't be produced at the scales required for commercial distribution and use.

Alone, none of these optimization challenges are insurmountable. But together they progressively increase the risk that a project's costs will balloon while also reducing the odds of success. These challenges can cause deadlines to slip, the wasting of critical resources, and, ultimately, project termination. Therefore bispecific platforms designed and engineered to reduce these risks can be instrumental to the field.

To this end, Twist Bioscience has partnered with Invenra to make the B-Body® bispecific platform widely available to the drug development community. The B-Body—a core protein framework designed to simplify and streamline the development and manufacturing of BsAbs—has been optimized to de-risk the bispecific drug development process in myriad ways. Together with Twist's infrastructure and expertise, B-Body bispecifics are poised to open the flood gates on BsAb therapeutic development*.

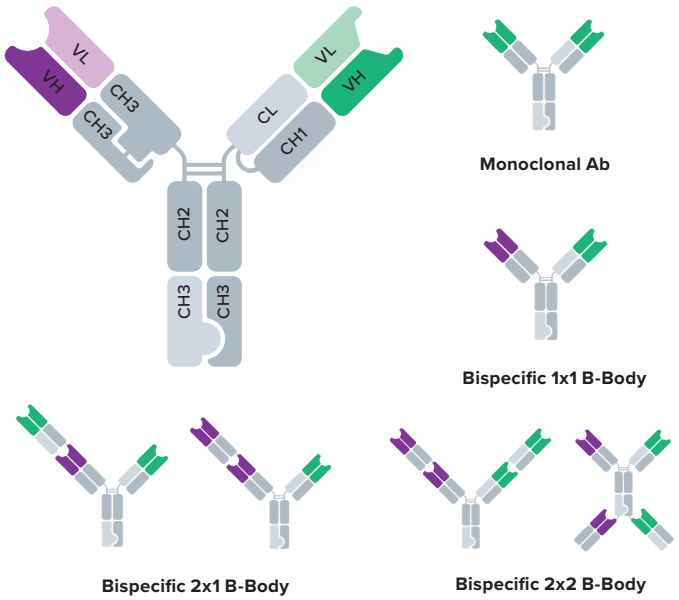
What is a B-Body?

At its core, a B-Body is a BsAb framework that resembles IgG antibodies in structure. Like monospecific IgGs, the standard B-Body construct contains two antigen binding domains (Fab's) and a crystallizable fragment (Fc) domain. However, unlike native antibodies which form symmetrical homodimers, a B-Body bispecific is an asymmetrical, heterotetrameric complex (**Figure 1**).

As indicated above, the B-Body construct has been carefully optimized to enable the robust pairing of two variant light chains with their cognate heavy chains. This enables efficient pairing, a reduction in product related contaminants, and the use of large-scale manufacturing processes comparable with those of standard mAb's. These optimizations include:

- **Tunable valency** is enabled by the B-Body bispecific design, such that additional Fabs can be added with varying configurations (**Figure 1B**.)
- **Robust Fc heterodimerization** driven by clinically validated Knob-in-Hole chemistry
- **Perfect light chain pairing** facilitated by an asymmetrical domain substitution, exchanging the canonical CH1-CL pairing for a modified CH3 dimer, as well as a Fab swap on the other side of the molecule
- **Minimal downstream process engineering** needed thanks to an engineered charge asymmetry between Fabs
- **High throughput one-step purification** enabled by the use of a single CH1 domain

Collectively, these optimizations turn the B-Body framework into a plug-and-play scaffold for BsAb development, one that can be used for large-scale screening of antibody binder pairings and one that is already tailored for large-scale manufacturing.



How does the B-Body framework de-risk BsAb development?

The B-Body framework de-risks the BsAb development process in various ways, whether its enabling plug-and-play testing of existing monoclonal antibody domains; improving the purity, yield and stability of the resulting BsAbs; or providing an industry validated construct that shows strong developability qualities.

An Industry Validated Tool

Perhaps the biggest risk that drug developers face is the unknown cost of using new, unproven tools. Even when innovators assure that their technology works, it's unclear how it will perform in real world conditions. When these tools don't work as expected, drug developers are set back in time and resources.

Although not well known, the B-Body construct has been used extensively in the drug discovery space for more than ten years. Between Invenra, partner organizations and Twist, more than 15,500 BsAbs have been built within the B-Body framework to date; and, importantly, one of these has advanced into clinical trials. In addition to being used for internal R&D needs, these thousands of BsAb molecules have been produced in service of more than 30 partnerships with drug development organizations. Similarly there are at least three CDMO's that have direct experience with the B-Body platform.

These BsAbs have included a diverse mixture of constructs, including those with either lambda or kappa light chains; more than 500 designs with Fc effector mutations; and over 50 germlines. While the majority of B-Body bispecifics made to date have followed the standard 1x1 valency, more than 1,000 BsAbs have been made with a 1x2 valency and >600 have included a 2x2 valency.

Put simply: This is not a new tool surrounded by unnecessary risk and uncertainty. B-Body bispecifics are a tried and true technology that has been validated by many drug discovery and development partners, and they continue to gain momentum globally.

Figure 1. Anatomy of a B-Body. (A) The B-Body has an IgG-like structure consisting of two Fab arms and a single Fc domain. Additional configurations and valency's can also be designed, as depicted here.

Easy, plug-and-play Fab and Fc testing

One early challenge in BsAb development comes when attempting to bring distinctly different antibody binding domains together in a single molecule. These domains may have been discovered using differing methodologies, frameworks, or isotypes. When combined, they can negatively influence one another, eroding the BsAb's overall function, expression level, and purity.

Additionally, developing a robust method to form desired pairingings in large quantities can be a challenge unto itself. Light-chain mispairing and unintended homodimer formation result in impurities that may reduce overall yield. Compensating for this may require bespoke purification workflows that are difficult to scale.

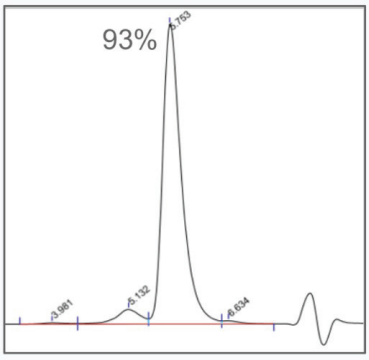
The B-Body architecture reduces these risks through multiple innovations. In addition to the clinically validated Knob-in-Hole Fc pairing technology, the B-Body construct is designed to avoid mispairing and homodimer formation by leveraging novel light chain paring solutions, which guide the highly efficient assembly of the B-Body bispecific. Notably, the charge asymmetry created between the two half-antibody arms within the B-Body enables especially robust charge related purification steps (e.g. ion exchange chromatography). This enables protein yields with up to 95% purity in high throughput settings as well as standard yields and purity with the use of standard mAb-like processes in cGMP manufacturing (**Figure 2**)

A significant advantage of the B-Body construct is that it allows for VL-VH pairs to be interchangeable—regardless of their source—without affecting the protein's developability. This is demonstrated by the data in Figure 3. To determine if BsAb expression is dependent on Fab source, 15 different Fabs (aligned with the Jain, et.al. paper) were reformatted to the B-Body framework in various pair orientations, with each Fab tested on both the knob and hole arms. The resulting protein's yield and purity levels were then measured in high throughput, using transient transfections and single-pass anti-CH1 purification.

Results from this study show that Fab identity, orientation (left vs right arm), and pairing had little impact on overall protein yield and assembly (as indicated by purity). Of the tested Fab pairings, a majority show over 90% purity with a yield of over 100mg/L (note: chain ratios of the individual polypeptides have been optimized for purity over yield in this high throughput format).

These results highlight the robustness and ability of the B-Body platform to be implemented in high-throughput workflows for the evaluation of 100's to 1000's of binding pairs and orientations.

A.



B.

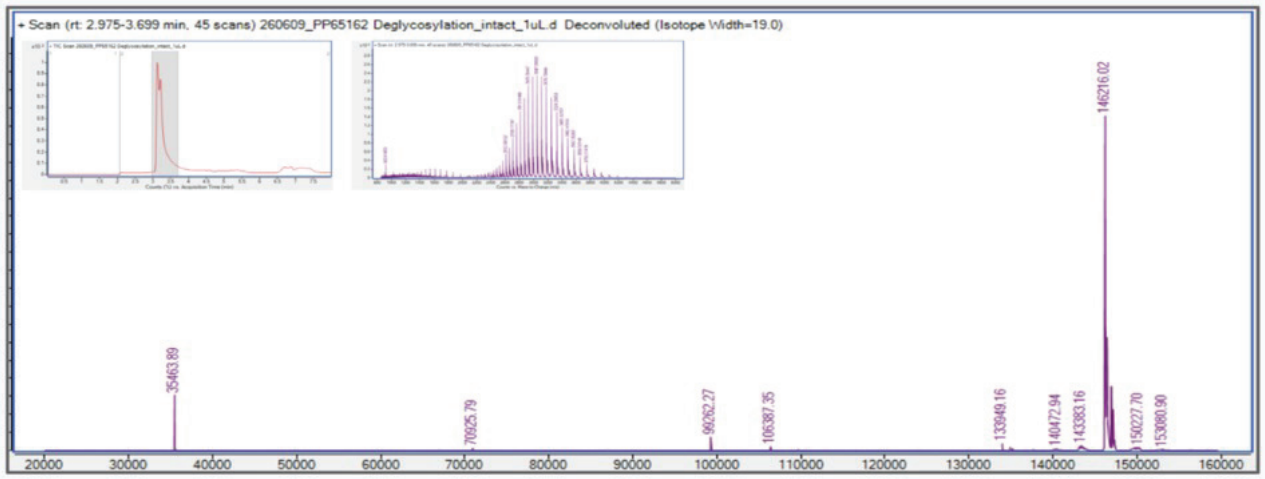


Figure 2. B-Body Architecture Eliminates Light Chain Mispairing and Homodimerization. Representative data from a CDMO demonstrating the purity of a B-Body bispecific antibody using (A) absolute size exclusion chromatography; and (B) liquid-chromatography mass spectrometry. In both graphs, the singular prominent peak represents a pure BsAb product.

Notably, it is sometimes desirable to modify the Fc domain for pharmacological or pharmacokinetic reasons. Here too, B-Body manufacturability appears unaffected by common Fc mutation variants. Table 1 lists common Fc mutations and the approximate number of B-Body constructs that have been made with these mutations to date.

Collectively, this data indicates that the B-Body framework's performance is robust across Fab constructs and is unaffected by commonly desired Fc mutations. Put another way, the B-Body framework enables the robust screening of 100's to 1000's of Fab pairings and Fc mutations with plug-and-play simplicity.

Consistently high yield and enduring stability

As indicated by Figure 3, B-Body bispecifics are, in part, optimized to produce a high protein yield. This is a critical attribute for therapeutics, which may be used by thousands of patients across the globe each year. When protein yield is low, drug manufacturers may need to compensate by expanding manufacturing infrastructure and resource investment to produce the needed quantities of drug product. It may not be economically or logistically feasible to do this, therefore a low protein yield increases the risk of high manufacturing costs and potential failure for otherwise promising therapeutic candidates.

Bispecific protein yield is influenced by several factors, including protein expression, amount of product related impurities, protein stability, and step yields of the downstream purification processes. In turn, these are all influenced by the protein's underlying amino acid sequence.

Fundamentally, the B-Body produces high BsAb titers from stable CHO cells that are commonly used in manufacturing settings. Additional substantiation can be found in a recent paper published by Invenra in the Journal of Pharmaceutical and Biomedical Analysis (JPBA).[3]

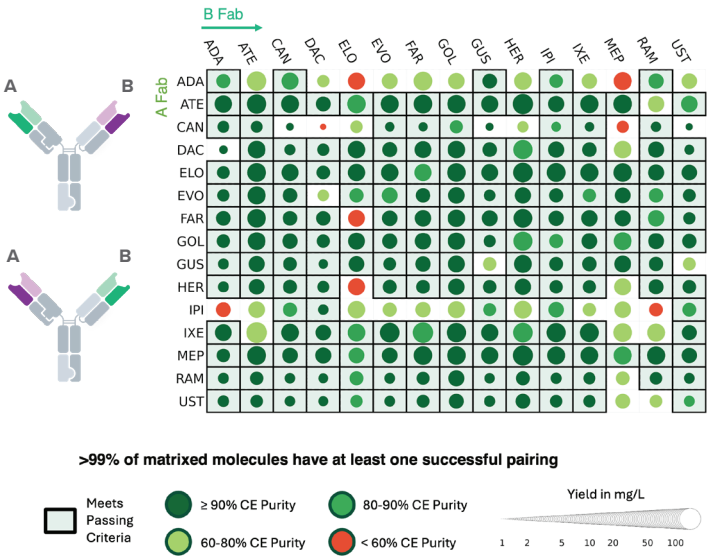


Figure 3. Plug and Play with the B-Body Framework. 15 different Fabs (aligned with the Jain, et.al. paper) were reformatted to the B-Body framework. Protein was produced using 1mL high throughput transient transfections and single-pass anti-CH1 purification. Results show the platform delivers consistent yield and purity across the 15x15 Fab pairing matrix.

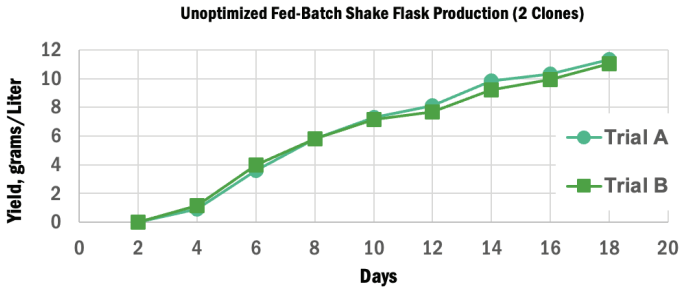


Figure 4. B-Body BsAb Yield. Representative example of B-Body bispecific antibody yield from a CDMO using stable CHO expression in fed-batch format. This graph shows increasing yield over time from two independent clones.

Fc Mutation	Residues Affected	Activity	Confirmed in B-Body® Format
LALA-PK	L234A, L235A, P329K	Silence effector function	Yes
Afucosylated	N297 glycan	Enhance effector function	Yes
GAALIE	G236A, A330L, I332E	Enhance effector function	Yes
LALA	L234A, L235A	Silence effector function	Yes
LALA-PG	L234A, L235A, P329G	Silence effector function	Yes
YTE	M252Y, S254T, T256E	Extend PK	Yes
LS	M482L, N434S	Extend PK	Yes
DLE	S239D, A330L, I332E	Enhance effector function	Yes
N297Q	N297	Remove glycosylation	Yes

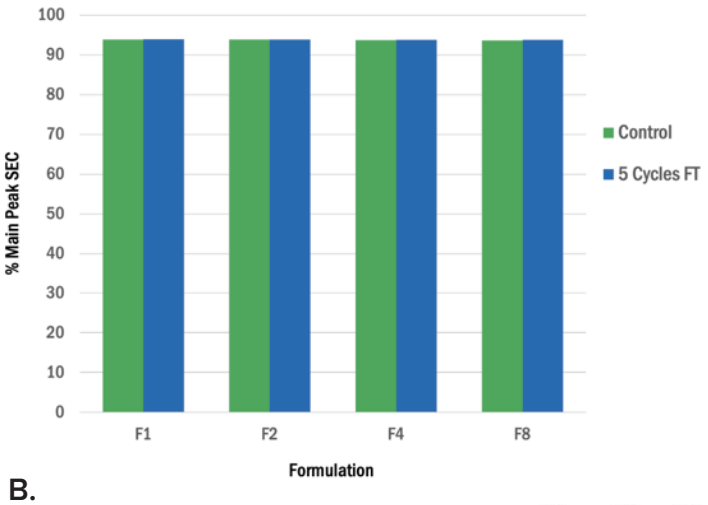
Table 1. List of common Fc mutations used in B-Body bispecifics.

Importantly, the biophysical stability of a therapeutic like a BsAb must go beyond the production facility, as they are likely to be shipped and stored for long periods of time. Stocks of therapeutics may undergo various forms of stress including temperature fluctuations, shear forces and agitation. To test if BsAbs are stable under these conditions, they are often tested for purity after repeated rounds of freeze-thaw cycles as well as months-long storage studies, both of which can be used as surrogates for stress conditions and used to calculate product shelf life (as well as other stability criterion).

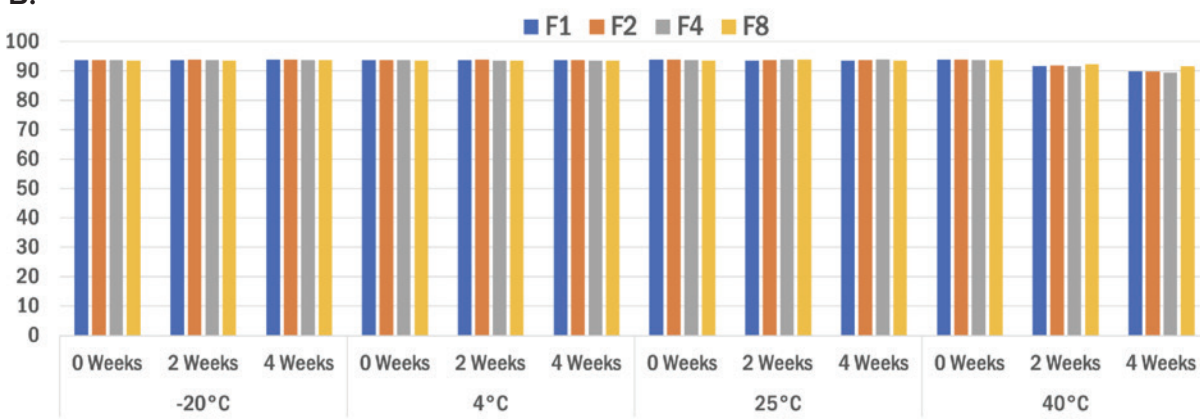
B-Body bispecifics have demonstrated robust stability across temperature ranges and freeze-thaw cycles, showing no reduction in purity when stored at -20°C, 4°C, and 25°C for four weeks (**Figure 5A**). When stored under extreme conditions (40°C) for the same time frame, stocks of BsAbs using a B-Body framework show marginal (3-4%) decreases in purity. Similarly, no reduction in protein purity was observed after five freeze-thaw cycles (**Figure 5B**).

Together, this data indicates that the B-Body format is stable and supports the long-term storage of BsAbs. Drug developers thus face less risk of producing a promising candidate that fails due to its frailty under working, shipping, and storage conditions.

A.



B.



Low immunogenicity and mAb-like pharmacokinetics

Engineered BsAbs come with the risk of eliciting immunogenic reactions in patients which can alter the safety and efficacy of the therapeutic in unanticipated ways. Immunogenicity may result when peptides contain non-human sequences (such as variable domains derived from murine antibodies) or novel engineered sequences that give rise to neo-epitopes. While the B-Body format is comprised of nearly 100% human sequences—containing very few amino acid substitutions—there is a formal possibility that immunogenic neo-epitopes have been generated.

To test this, B-Body bispecifics have been generated with Trastuzumab variable domains (forming a monospecific B-Body). In vitro immunogenicity of this monospecific B-Body was compared with the parental Trastuzumab mAb molecule. In this context, the immunogenic potential of the Trastuzumab B-Body has been shown to be equivalent to the monoclonal Trastuzumab antibody (**Figure 6**).

The safety and efficacy of any therapeutic is partly determined by its pharmacokinetics. Fc containing biologics, including many BsAbs, tend to have half-lives akin to monoclonal antibodies. As expected with the B-Body framework, they too follow this trend, showing a pharmacokinetic profile in primate models with a half life of ~10-12 days, very similar to standard monoclonal mAb's (**Figure 7**).

Figure 5. Stability of B-Body Bispecifics. (A) B-Body formulation purity is maintained across five freeze-thaw (FT) cycles. **(B)** B-Body bispecifics demonstrate robust stability, as measured in purity (Y-axis) across temperature conditions over four weeks, with 3–4% decrease in purity observed only after 4 weeks of storage under 40 degree celsius conditions.

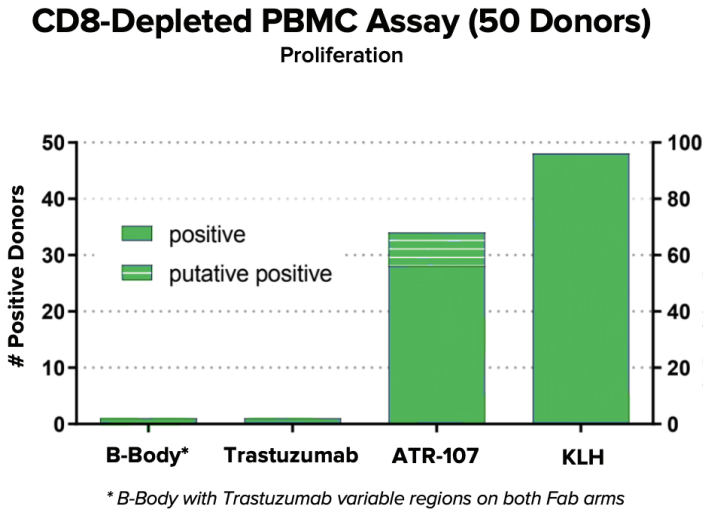


Figure 6. Immunogenicity of B-Body bispecific construct. Data represents the results from an in vitro immune activation index assay, which combines multiple pro-immunogenic readouts to determine a molecule's immunogenicity. Here, Trastuzumab variable domains were transferred to a B-Body format and demonstrated no increase in immunogenicity compared to the parental Trastuzumab antibody. ATR-107 = a positive control antibody known to elicit an immunogenic response. KLH = keyhole limpet hemocyanin protein. Activation index based on multiple pro-immunogenic readouts flow cytometry, proliferation, etc.

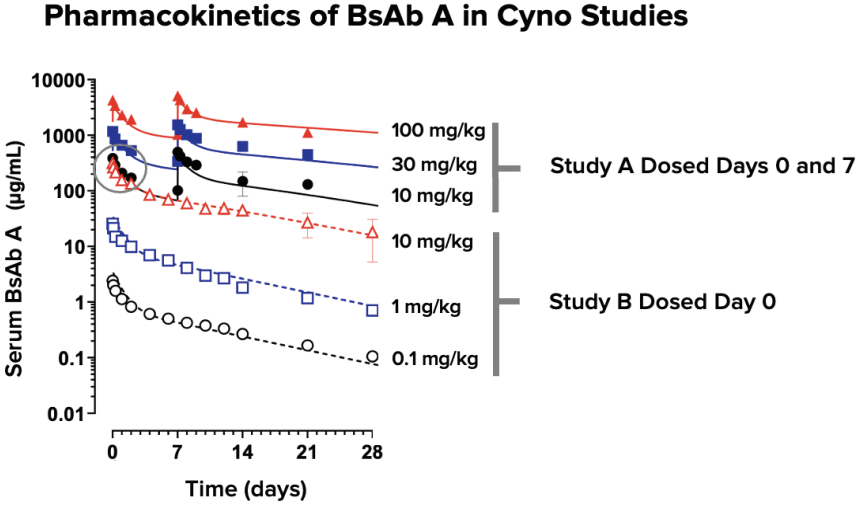


Figure 7. B-Body Pharmacokinetics. Bispecifics show MAb-like pharmacokinetic profile in cynomolgus primate studies.

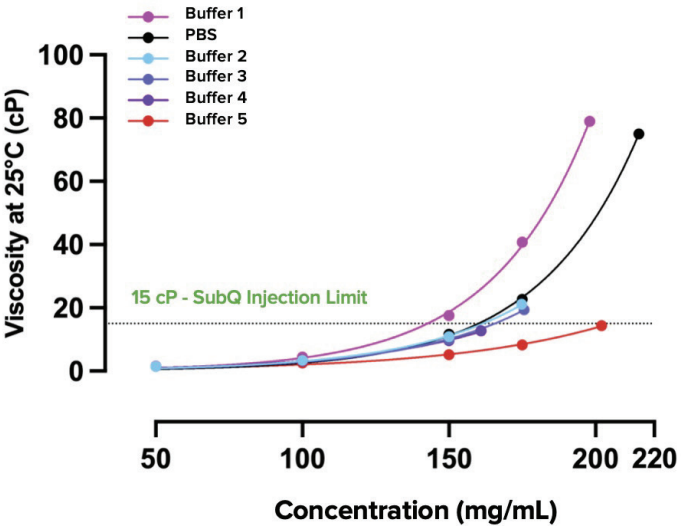


Figure 8. B-Body Formulation Compatibility With Subcutaneous Injection. Representative results from a CDMO-performed buffer formulation screen. Buffers 1-5 are standard therapeutic antibody buffers using compendial excipients. SubQ = subcutaneous.

High concentration formulation compatible

The administration of most antibody based therapeutics is typically via a slow infusion. This is often determined to be required either due to potential adverse events (injection site immunological reactions) or simply that the molecule cannot be formulated at a high enough concentration with sufficiently low viscosity to be compatible with subcutaneous delivery routes. Whilst the adverse events are often the result of biology, the formulation issues tend to be a consequence of the antibody formats. With respect to the later, bispecific antibodies complicate this in that they are comprised of two variable domains which may interact in unexpected ways, leading to aggregation and other biophysical issues that limit their ability to be formulated at the high concentrations needed for low volume, syringe-based delivery.

Figure 8 demonstrates the inherent solubility provided by the B-Body format. In a simple buffer screen (performed by CDMO using B-Body bispecifics), a B-Body BsAb was formulated up to 200mg/mL without exceeding the viscosity limit (15cP) for subcutaneous injection. The BsAb could even be formulated in PBS up to ~150mg/mL without significant viscosity limitations. This result will vary by the different binding domains inserted into the B-Body, but the unique charge asymmetry within the B-Body is likely to provide drug developers with a unique opportunity to attain high concentration formulas with most B-Body bispecifics.

Twist Bioscience and the new era of BsAb development

Engineered by Invenra, the B-Body platform has proven to be a valuable asset for drug developers interested in BsAbs. The optimized framework greatly simplifies the process of combining Fabs from various origins, enabling the highly efficient formation of desired BsAb constructs in high-throughput formats. Additionally, the platform robustly delivers high purity and yield of BsAb products using standard mAb manufacturing platforms. B-Body molecules also display the robust biophysical stability that's needed for therapeutic manufacturing, formulation, use and storage.

In summary, the B-Body platform greatly de-risks BsAb development by providing clear improvements in the process from early discovery through to clinical manufacturing. This means that researchers can focus on rapidly screening Fab pairings in a chemistry, manufacturing, and controls (CMC)-ready format with minimal risk.

With its automation, infrastructure, and global reach, Twist Bioscience is democratizing access to the B-Body bispecifics platform in a way that no others can.

References

1. Antibody Society. Database for Therapeutic Antibodies: YAbS. Antibodysociety.org. 2026. Accessed September 8, 2026. https://db.antibodysociety.org/

2. National Library of Medicine. Drug Trials Involving Bispecific Antibodies | Search Results. Clinicaltrials.gov. 2026. Accessed September 8, 2026. https://clinicaltrials.gov/search?intr=%22bispecific%20antibody%22&viewType=Table&aggFilters=status:act%20rec%20not%20afm%20ava%20enr%20unk

3. Chen X, Gawdzik J, Chen H, et al. Forced degradation studies reveal favorable stability of bispecific antibodies generated using the B-Body® platform. Journal of Pharmaceutical and Biomedical Analysis. 2027;282:117716. doi:10.1016/j.jpba.2026.117716

**For Research Use Only. Not for use in diagnostic procedures. Data generated by Invenra, Inc in partnership with Twist Bioscience. Results may vary based on project specifications.*