
Harvest[®] Technologies BMAC[®] Data Summary

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Lille Tidwell, PhD^{*}, Nésa-Maria Anglin^{*}, John Genereux^{*} and Sherwin Keyv, MD^{**}

^{*}Harvest Technologies, Inc., Plymouth, MA

^{**}Assoc. Prof. Pediatrics, Dept. Transfusion Service Emeritus, Children's Hospital, Harvard Medical School, Blood Research Institute, Boston, MA

Background:

Since 2011, Harvest[®] has collected data from over 2,200 customers using the Harvest[®] SmartPRep[®] system for Bone Marrow Aspirate Concentrate (BMAC[®]). With a recent Institutional Review Board approval, Harvest[®] can now present data from August 2011 to December 2013 and provide very interesting summaries that will help to understand the usage of concentrated bone marrow in the market. This data represents what has been reported through the BMAC[®] sampling program, therefore, it serves as a very good indicator for the entire customer base. Harvest[®] can also consider how this data may represent BMAC[®] in the wider orthobiologics market.¹

The goal of concentrating bone marrow is to achieve a regenerative autologous biologic. The bone marrow cells in our bodies naturally respond to injury by responding to chemo-attractant signals released at the site of injury.² Numerous papers demonstrate the use of autologous concentrated bone marrow as a safe biologic approach in a variety of clinical applications.^{3,4,5}

Harvest[®] originally established the BMAC[®] sampling program as a way to report the actual cell counts in a sample of bone marrow aspiration and the concentrate to surgeons who were not as familiar with bone marrow aspiration technique. The cell counts typically obtained when concentrating bone marrow aspirate (BMA) using the Harvest[®] SmartPRep[®] system are comparable to the first papers demonstrating the use of concentrated bone marrow for orthopaedic applications.^{6,7} Harvest[®] is the only company that provides this type of sampling program to its customers and can now provide follow-up data from a large data set. This summary will be used to educate customers and sales teams to recognize that the quality of a bone marrow concentrate system is crucial in the decision making process when choosing a biologic.

Results:

The BMAC[®] Marrow Sample Data Forms are filled out in the field by sales representatives, then the pre and post concentration samples are collected and shipped overnight at ambient temperature for analysis. The samples are analyzed for Complete Blood Count and the resultant data is entered into a database by laboratory staff. A summary report is created and sent back to the technical field sales representative and the physician. Since these forms are hand-written and filled out in the field, it is expected that the forms may not be complete or contain errors. Therefore, the sample size for each parameter is unique and will be identified as presented.

The original raw data was sorted according to lab date. Original BMAC[®] Marrow forms were reviewed and corrections were made if possible. This provided 2,013 samples that could be included in the initial review of the following parameters that could teach Harvest about the marketplace and system performance: age, gender, surgical sites, and SmartPRep[®] system performance (Total Nucleated Cells (TNC), Platelets (PLT), % Yield, Concentration factor, and Hematocrit Levels).

Age and Gender

From 2,013 samples the majority, 77% (1,550), of data represents patients over the age of 40, while 20% (408) represents patients 40 years of age or younger, and the remaining 3% (55) represents inconclusive data. It was determined that 52% (1,051) samples represented females, 47% (951) samples represented males and 1% (11) represented inconclusive data. This data demonstrates that the majority of the patients receiving Harvest[®] BMAC[®] are over 40 years of age with a slightly higher representation of female patients.

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Surgical Sites

From a total number of 2,013 patient samples, there were 3 patients that had more than one surgical site at one time, making the total number of surgical delivery sites 2,016. Based on reading the comments on the BMAC[®] Marrow Sample Data Forms where sales representatives were asked to report the type of procedure, there were 10 different surgical sites: Spine, Hip, Knee, Shoulder, Lower Limb, Upper Limb, Foot & Ankle, Hand & Wrist, Oral Maxillary Facial (OMF), Eye and Other **Figure 1**). In all 2,013 patients, the iliac crest was the primary aspiration site. The tibia was aspirated as an additional site in 5 patients and vertebral bodies through pedicles were aspirated as an additional site in 60 patients.

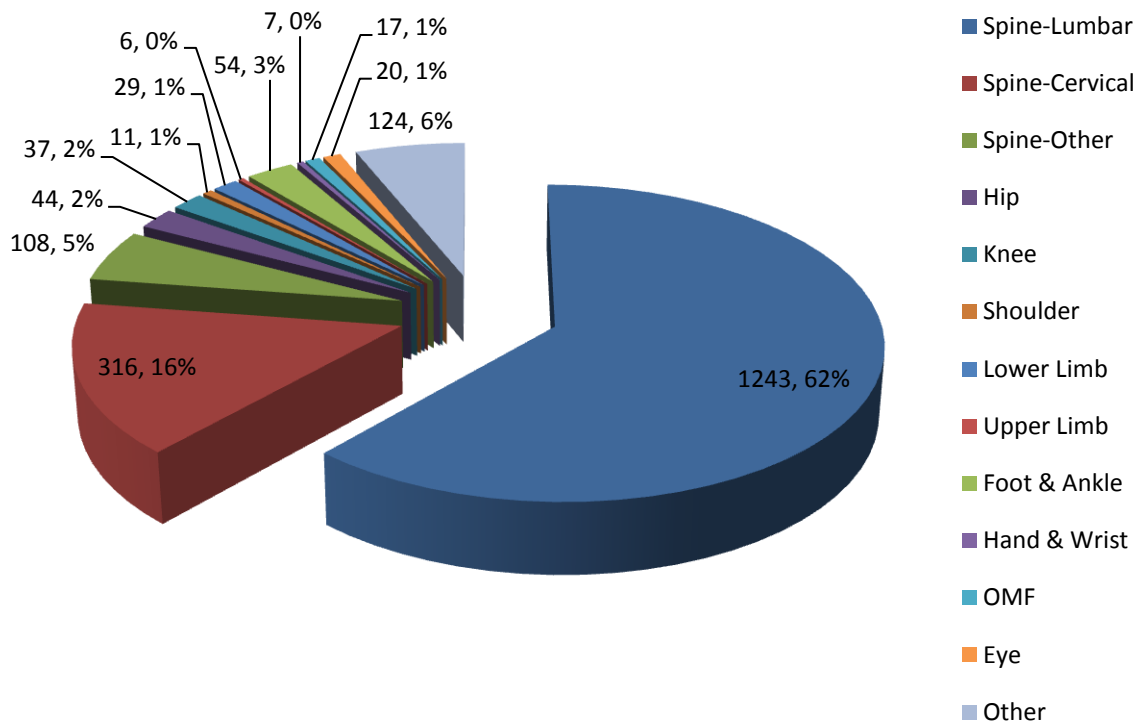


Figure 1: Summary of the types of surgical sites reported in data collected from August 2011 to December 2013

Spine represented a total of 83% and was subcategorized into “Spine-Lumbar” with 62%, “Spine-Cervical” with 16% and “Spine-Other” with 5% which included thoracic (generally scoliosis surgeries that ranged from T3 to sacrum) and general descriptions that indicated the surgery was spine but no specifics were provided.

The next three categories of Hip, Knee and Shoulder could be joined together to represent Joints, a total of 5% of surgeries, where each category represented 2%, 2%, and 1% respectively.

Lower Limbs (1%) and Upper Limbs (< 1%) represented less than 2% of surgeries and included descriptions of non-unions, fractures, tumors, hardware removal, revision or tendon repair.

Foot & Ankle (3%) represented a fairly large group and included fusions, tendon repair or lengthening, arthrodesis, non-unions, hardware removal, revisions, osteochondral lesions, osteotomies, fractures, and so forth. Hand & Wrist represented a group of less than 1% and primarily included hemitrapeziectomies with carpometacarpal arthroscopy.

Oral Maxillary Facial represented 1% of surgeries and included procedures such as bone grafts to the maxilla and mandibular reconstruction.

There is one physician using BMAC[®] for ophthalmic injections which represented 1% of surgeries. This was a surprising find and perhaps represents a new market area⁸.

The “Other” category represented 6% surgeries where the anatomical location could not be determined due to insufficient notes. We hope to inquire with the sales team for clarification and that the forms will be more complete in the future.

Biological Parameters

There are several methods of isolating or concentrating blood and bone marrow, but the buffy coat method is the most commonly used. The buffy coat is described as the fraction of an anti-coagulated blood or bone marrow sample that contains most of the white blood cells and platelets following density gradient centrifugation. The Harvest[®] SmartPrep[®] BMAC[®] system concentrates 100% bone marrow aspirate, not a combination of marrow and blood, using the buffy coat method with a proprietary floating density shelf that captures a high percentage of the cells in the buffy coat, including platelets that are found within and above the buffy coat. The captured cells and platelets are then measured by a commonly used Complete Blood Count (CBC) analysis with an automated cell counter (Coulter AcT-diff2). After considering certain variables, inclusion criteria were applied to the 2,013 samples before we analyzed the data further. These criteria excluded the sample data based on the following;

- 1) Age and Gender not reported
- 2) Biologic parameters with N/A, blanks, or inconsistencies
- 3) BMA and BMAC volumes that followed the Instruction For Use
- 4) Samples that had greater than 100% Yield for Total Nucleated Cells or Platelets
- 5) Hematocrit % greater than 100

After applying these exclusion criteria 952 samples were then categorized according to the different configurations of BMAC[®] kits. The biological parameters are summarized in **Table 1**.

Table 1: Summary of the BMAC data collected from August 2011 to December 2013

Kit Used	Sample Number	BMA BMAC	Total Nucleated Cells (X 10 ⁶ / mL)				Platelets (X 10 ³ / μL)				Hematocrit %		
			Average	±SD	Median	Average Conc. Factor	Average	±SD	Median	Average Conc. Factor	Average	±SD	Median
240mL	2	BMA	19.7	2.4	19.7		108	23	108		31.2	1.2	30.5
		BMAC	83.8	12.7	83.8	4.2 ± 0.1	508	337	508	4.5 ± 2.2	35.7	5.9	39.1
180mL	19	BMA	25.1	12.9	19.6		107	55	102		31.2	5.2	30.9
		BMAC	89.5	31.2	80.6	3.9 ± 1.1	400	213	443	4.1 ± 1.8	24.9	10.5	22.4
120-20mL	215	BMA	21.7	11.2	19.5		122	129	107		32.5	4.8	32.5
		BMAC	83.3	46.6	73.1	3.9 ± 1.0	467	356	408	3.9 ± 1.4	29	11.6	27.4
60-7/10mL	561	BMA	21.8	12.2	19.6		117	70	106		32.9	14.9	32.1
		BMAC	84.5	49.1	74.2	3.9 ± 1.2	457	329	407	3.8 ± 1.6	29	11.6	26.4
30-4mL	155	BMA	20.1	9.1	18.5		109	71	95		32.4	5.1	32.4
		BMAC	80.7	46.7	69.3	4.0 ± 1.3	450	336	378	4.1 ± 2.0	29.3	12.4	26.6
TOTAL	952												
Avg BMA			21.7	9.6	19.4		112	70	104		32.0	6.2	31.7
Avg BMAC			84.4	37.3	76.2		456	314	429		29.6	10.4	28.4

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Total Nucleated Cell Counts and Concentration Factor

The total nucleated cell (TNC) count provides an accepted method to measure the efficiency of a bone marrow concentration system. The Harvest® SmartPrep® system provides a consistent and high nucleated cell count compared to other systems on the market. The data in **Table 1** demonstrates that the average TNC, for BMA samples, was 21.7 ± 9.6 nucleated cells $\times 10^6/\text{mL}$ with a median of 19.4 nucleated cells $\times 10^6/\text{mL}$. The average TNC, for BMAC® samples, was of 84.4 ± 37.3 nucleated cells $\times 10^6/\text{mL}$ with a median of 76.2 nucleated cells $\times 10^6/\text{mL}$. The concentration factor was approximately a 4 fold concentration compared to the baseline aspirate. The collective data was also subcategorized according to the BMAC® kit configurations. The most common kit used is the BMAC2 60-01 where 60 mL of aspirate can be concentrated to either 7 or 10 mL. The average cell counts in 561 aspirate and concentrate samples that were processed using the BMAC2 60-01 kit are almost identical to the overall average.

Platelet Counts and Concentration Factor

Platelet concentration is important when considering the composition of concentrated bone marrow. Sometimes it is forgotten that the yellow plasma just above the white buffy coat layer actually has concentrated platelets which will release needed growth factors for the bone marrow cells in the buffy coat layer to function properly, such as migration and proliferation. For this reason, it is very important to not remove volumes below what is described in the Instruction For Use protocol in the BMAC® kits. The Harvest® SmartPrep® BMAC® system concentrates platelets from an average of $112 \pm 70 \times 10^3/\mu\text{L}$ with a median of $104 \times 10^3/\mu\text{L}$ in BMA to $456 \pm 314 \times 10^3/\mu\text{L}$ with a median of $429 \times 10^3/\mu\text{L}$ in BMAC®. The concentration factor was approximately a 4 fold concentration compared to the baseline aspirate. Similar to the cell counts, the average platelet counts in 561 pre and post processed samples using the BMAC2 60-01 kit are almost identical to the overall average.

Hematocrit Levels

Hematocrit is a measure of the percentage of whole blood (or bone marrow) that is made up of red blood cells. The desire in concentrating BMA is to maintain a Hematocrit below 30% which has minimal increase in cellular viscosity.⁹ The average Hematocrit level in BMA was 32% and the BMAC® had an average of 29.6%. There is consistency for average and median Hematocrit percentage amongst all the different types of kits used. It should be emphasized that this data comes from volumes in aspirate and concentrate that follow the guidelines in the Instruction For Use. Variation was observed in the aspirate and concentrate volumes which can be explained by a number of different factors. In order to achieve the best results for total cell counts, platelets, and hematocrit levels it is important to stay within the guidelines of the Instruction For Use protocol.

Discussion:

The initial evaluation of this data demonstrates that the age and gender distribution matched what was expected from marketing research; that the majority of patients receiving BMAC® were over the age of 40. There was not a significant difference between male or female. Spine procedures represents the largest amount of BMAC® sample data (83%) for Harvest® which is similar to internal sales data. Interestingly, it appears that Cervical Spine procedures using BMAC® may be increasing, and the Foot/Ankle, Joint, and Ophthalmologic markets may represent new areas of growth for Harvest®.

Across the literature pertaining to the use of bone marrow derived cells or aspirate/concentrate, the primary parameter that can be consistently compared are nucleated cell counts, also referred to as total nucleated cell counts. Muschler et al.¹⁰ reported 56 nucleated cells $\times 10^6/\text{mL}$ in a graft of iliac crest bone and 19 nucleated cells $\times 10^6/\text{mL}$ in bone marrow aspiration. Using 25 normal bone marrow samples, Hermann et al.¹¹ used the Harvest® SmartPrep® BMAC® system and reported BMA to contain 23.1 ± 5 nucleated cells $\times 10^6/\text{mL}$ and 89.1 ± 8 nucleated cells $\times 10^6/\text{mL}$ in the concentrate. There are two more publications this year that used the Harvest® SmartPrep® BMAC® system and

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provided nucleated cell counts that can also be compared with this current data analysis. Hegde et al.¹² reported two separate sets of data where BMA contained 18.62 ± 12.16 or 16.62 ± 8.42 nucleated cells $\times 10^6/\text{mL}$ and BMAC[®] contained 101.48 ± 64.13 or 90.80 ± 48.90 nucleated cells $\times 10^6/\text{mL}$. Johnson¹³ reported 20.5 ± 10.0 nucleated cells $\times 10^6/\text{mL}$ contained in BMA and 66.6 ± 43.9 nucleated cells $\times 10^6/\text{mL}$ contained in BMAC[®]. The Harvest[®] BMAC[®] sampling program data demonstrates an average of 21.7 ± 9.6 nucleated cells $\times 10^6/\text{mL}$ in BMA and 84.4 ± 37.3 nucleated cells $\times 10^6/\text{mL}$ in BMAC[®], which is similar to what has been published by the two independent orthopaedic teams mentioned above as well as what was observed by Muschler et al. and Hermann et al.

While the platelet counts are not a focused-on parameter in a bone marrow aspirate concentrate preparation, the fact that the concentration factors were consistent for total nucleated cells and platelets in BMAC[®] demonstrates consistency and overall performance of the Harvest[®] SmartPRep[®] system.

Another indicator of system performance is the fact that average Hematocrit levels did not increase above baseline when concentrating bone marrow aspirate. As mentioned above, maintaining a Hematocrit below 30% has minimal increase in cellular viscosity.⁹ This can be very important when surgeons want to use concentrated bone marrow for percutaneous injections without activating clot formation.

The Harvest[®] BMAC[®] sampling program has provided Harvest[®] with field data from customers who are non-hematologists and have not historically aspirated bone marrow, yet the nucleated cell and platelet counts are very similar to publications and data produced by a third party (BioScience Research Associates) which was submitted to the FDA for the Harvest[®] SmartPRep[®] 510K approval in 2006. Thus, the Harvest[®] SmartPRep[®] system has consistently provided the highest nucleated cell and platelet counts than any other competitive product on the market for more than 8 years to a large customer distribution.

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