MAF 314® INSTRUCTION MANUAL

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A brief history of MAF 314® by Prof. Marco Ruggiero and Dr. Stefania Pacini

We have been working on vitamin D and its receptor (vitamin D receptor, VDR) for many years, identifying the VDR gene profiles associated with a number of physiological and pathological conditions. Vitamin D, VDR and vitamin D binding protein are called “vitamin D axis”. (You can find our papers in www.pubmed.org)

Therefore, when Prof. Yamamoto in 2009 published his paper claiming that GcMAF [Gc protein-derived macrophage activating factor] eradicated HIV without antiretroviral therapy (ART), it was only natural for us to direct our attention to GcMAF because it belongs to the vitamin D axis in that GcMAF derives from vitamin D binding protein, also known as Gc-protein.

We were also attracted by the possibility to demonstrate in the laboratory that the famous words of Prof. Montagnier—“…you will get rid of the virus in a few week if you have a good immune system”—were true. Not that we doubted his words, but we knew that his words were based upon his clinical observations and not on actual experiments performed in the laboratory. This approach, often referred to as “immunotherapy”, would shift the focus from the fight against the virus to the effort to re-establish, or empower, an immune system made deficient by a number of different causes, probably different for each individual.

Because of these considerations, we thought that immunotherapy with GcMAF would be a good object of study. There was also another consideration. AIDS diagnosis and the decision to initiate ART are based on CD4 cell count. Therefore, we reasoned, if we are able to keep them high, HIV+ people, in addition to staying healthy, will avoid prescription of ART and will not be classified as having AIDS.

We had just recently read a very interesting article by Reid et al. (quoted in our presentation at the 6th International Aids Society Conference on HIV Pathogenesis, Treatment and Prevention) where he demonstrated that a probiotic yogurt was able to rise CD4 to an extent comparable to that of ART, obviously without side effects and practically at no cost. In fact,
it was produced by local women in low-income communities in Tanzania. In other words, locally-produced probiotic yogurt produced the same effects of ART.

Reid et al. did not provide a molecular mechanism underlying the observed effects on CD4 cell count. We thought that bacteria contained in their probiotic yogurt could have converted some Gc-protein that is present in low concentration in milk into active GcMAF. Thus, administration of Reid’s probiotic yogurt might have mimicked Yamamoto’s administration of GcMAF.

Based on these premises, we then studied a way to have certain strains of bacteria convert high concentration of Gc-protein into GcMAF and we developed MAF 314®. This was not easy at all and the number 314 indicates the number of different combinations/experiments we had to perform before obtaining the right conditions. This is definitely not something that you could perform in your home kitchen! Then, we thought that it could have been cooler to call it 314 mimicking the p number.

We also took advantage of the deep knowledge of human anatomy and histology of Dr. Stefania Pacini who is teacher of human anatomy at the Faculty of Medicine of the University of Firenze. In fact, she let us know that there was no need to inject MAF 314® (an impossible task) if we wished to stimulate the immune system. The existence of the mucosa-associated lymphoid tissue (MALT) that comprises macrophages is now widely recognized and activated macrophages can re-circulate between mucosae, blood, tissues, and back to mucosas. At this point the only remaining obstacle was the acid environment of the stomach that could have digested the GcMAF presented in MAF 314®. This problem also was solved by Dr. Pacini’s knowledge of human anatomy. In fact, in the Waldeyer’s tonsilar ring there is an abundance of macrophages that can be directly stimulated by the GcMAF present in MAF 314®. Not only that, we developed a way to have most MAF 314® pass intact the gastric environment and arrive intact (for the most part) in the lower intestine where the gut microbiota (now conceived as a virtual organ) exerts its actions.
In fact, MAF 314® is not only a GeMAF-enriched yogurt. Based on Reid’s publications and successes, MAF 314® was developed to re-establish a healthy gut microbiota, very similar to that of newborn mammals. A sort of reset of the gut microbiota. Again, this achievement was not easy.

At that point, we began experimenting on ourselves as true scientists of the old days used to do. You can see the results of this “trial” in IAS2011. Immediately thereafter, we gave MAF 314® to a couple of friends with different pathologic conditions and we are now monitoring their situation. By the way, MAF 314® has a good taste and people who tried it found it good. We are collecting a series of unexpected positive side-effects that we believe are due to the lucky combination of immune stimulation and re-establishment of a healthy gut microbiota. Many of these effects are just sensations difficult to quantify in scientific terms, but they are nevertheless very welcomed by those experiencing them.

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About the Authors

Stefania Pacini is a researcher and teacher of human anatomy at the University of Firenze, Italy. She has a degree in Biological Sciences and one in Medicine and Surgery. She worked as visiting scientist at the Laboratory of Cellular and Molecular Biology (Chief: Dr. S. A. Aaronson) of the National Cancer Institute (NIH, Bethesda, Maryland) on signal transduction and oncogenes. Her current researches focus on cell morphology and as well as on genetic analysis of polymorphic genes. She teaches in the faculty of Medicine and Surgery of the University of Firenze, Italy. She is the author of more than 80 scientific papers.

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Marco Ruggiero, MD, PhD, is a professor of Molecular Biology at the University of Firenze, Italy. He has a specialization in clinical radiology and served as Lieutenant Medical Officer in the Italian Army. In 1984-86 he worked on signal transduction and protease inhibitors as a post-doctoral fellow at Burroughs Wellcome Co. (Research Triangle Park, NC) with Drs. Cuatrecasas and Lapetina. One of his papers on protease inhibitors was presented to the Proceedings of the National Academy of Sciences by Nobel laureate Sir John Vane. Subsequently he worked as visiting scientist at the Laboratory of Cellular and Molecular Biology (Chief: Dr. S. A. Aaronson) of the National Cancer Institute (NIH, Bethesda, Maryland); his research was focussed on oncogenes and signal transduction. In 1992, he moved back to Firenze, Italy, where now he teaches in the Faculties of Mathematical, Physical and Natural Sciences, Medicine and Surgery, and Engineering. He has been the tutor of many students preparing Bachelor or PhD. theses, several of which have been on AIDS with particular emphasis on the non-viral origin of the disease. He is the author of more than 100 scientific papers in journals such as Science, PNAS or Oncogene, and he has been recently appointed Author in Chief of the “Springer Reference Live: Cancer”.

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KITCHEN TOOLS REQUIRED FOR THE PREPARATION OF MAF 314®

(for one week)

(A) One pot for boiling two liters of milk; steel and non-stick pots are appropriate

(B) One glass bowl of at least one liter capacity

(C) One measuring cup of at least one liter capacity

(D) One regular steel tablespoon

(E) One plastic or steel tablespoon with a long handle
(F) One large plastic or steel ladle

(G) A medium-mesh strainer

(H) One glass jar (at least one liter capacity) with lid

(I) One glass jar (at least one liter and half capacity) with lid

(J) A yogurt maker. Note: The yogurt maker will typically have its own set of cups

(K) 14 little jars (capacity: 100 ml each) 7 little jars (capacity 125 ml each for one person)
(L) A digital food thermometer

(M) One measuring scoop

(N) Several rolls of plain paper towels

(O) Aluminum foil

(P) Sticky labels

(Q) Cleaning sponge

(R) Liquid dishwashing soap
INGREDIENTS REQUIRED FOR THE PREPARATION OF MAF 314®

(to make one week's supply starting from scratch)

1. **Two liters of whole high quality pasteurized cow milk**, without any of the following added: preservatives, vitamins (A and D), hormones, or other chemical substances. Raw milk from certified producers can also be used as well as goat or sheep milk. Preferably, do not use UHT milk. **DO NOT USE**: rice milk, soy milk, almond milk, or lactose free milk.

   - MAF 314® starter #1

   - MAF 314® starter #2

   - MAF 314® probiotic #3

   - **One bottle** (125 ml) of **bovine colostrum** (e.g., from LR Health and Beauty). Colostrum from other certified producers as well as raw colostrums is also suitable.
HOW TO PREPARE MAF 314®
(for one week)

BEFORE STARTING

- Choose a wide and well-lit working surface. Ensure it has a nearby electrical outlet for the yogurt maker. Clean it carefully with hot water and liquid dishwashing, then rinse and dry it well with paper towels.

- Wash each time, with hot water and liquid dishwashing soap and before and after each use, all the tools required for the preparation of MAF 314®: pot, ladle, bowl, jars, etc… Rinse them very well, then dry them carefully with paper towels—and never use the same paper towels more than once. Some utensils (spoons, ladle and measuring cup) will be used several times in the same day to prepare the different compounds. In order to prevent contamination, please wash them well with hot water after each individual use, rinse and dry them with paper towels.
- Use the kitchen tools required for the preparation of MAF 314® ONLY for this purpose—DO NOT USE them for cooking other foods - and, after having cleaned them, store them separately from the others kitchen tools commonly used for cooking. Do not cook other foods while preparing MAF 314® since contamination might occur.

- Some products will be left out at “room temperature”. Since different people prefer different temperatures at their homes, understand that for the purposes of making MAF 314®, “room temperature” means 26 – 30° C.
DAY ONE

Preparation of Compound #1

1. Heat the milk (two liters) until boiling. Stir every few minutes with the plastic or steel tablespoon. When the milk boils move the pot away from the cooking stove and let the temperature cool down to 40 – 42° C. Check the temperature with the thermometer. Keep the thermometer clean; clean it after each use. If the temperature is slightly lower than 40°C, no problems occur. However, if the temperature is significantly lower than 40°C, it is possible to warm again the milk for a few minutes without boiling it.

2. If present, remove the dense layer on the milk surface using the spoon. This will allow more rapid cooling and guarantees a more homogeneous product.

3. While you are waiting for the milk to cool down, place the yogurt maker on the working surface. Be sure that its little jars are clean (if necessary wash them with hot water and liquid dishwashing, then rinse and dry them carefully with paper towels). Place the little jars (open, without their lids) into the yogurt maker.

4. Close carefully the yogurt maker with its own transparent lid and pre-warm the jars so that they will be at the right temperature when you are ready for the next steps.

5. When the temperature of the milk is between 40 - 42° C, transfer one liter of milk in the measuring cup. Add to this quart of milk, 1 bag of starter #1 and stir carefully with the tablespoon.

This first liter of milk will become the first component of MAF 314®, that is compound #1.

6. In order to complete preparation of compound #1, split this liter of milk in the 7 little jars that were previously placed and pre-warmed into the yogurt maker. Close well the yogurt maker with its own transparent lid (but leave open the little jars). Wait for 9 hours.
7. After 9 hours, the product in the small jars will appear dense and thick. Now open the lid of the yogurt maker, collect all the little jars, close them well with their own lids and place them into the refrigerator for 12 hours. It is preferable to perform this procedure at the end of the 9 hours. However, if this is not feasible, the jars can remain in the switched off yogurt maker for a few hours before being collected and placed in the refrigerator.

You have now obtained the MAF 314® compound #1 in the yogurt maker cups

Preparation of compound #2

1. The other liter of milk (measure it by the measuring cup) that had been previously boiled and was not used to prepare compound #1, has to be further cooled down until reaching room temperature (26 - 30° Celsius). This second liter will be used to prepare compound #2. If some milk has been lost during the steps of the preparation, boil some other milk and cool it down to 26-30°C in order to have 1 full liter for the preparation of compound #2.

2. When you are sure that the temperature is correct (check it using the thermometer), take a big spoon of milk with the ladle and place it into the glass bowl. Carefully open 1 bag of starter# 2 and add all its content to this small volume of milk, stir carefully with the table spoon to dissolve all the lumps. Add the remaining milk up to reach the volume of one liter and stir carefully with a clean tablespoon

3. Loosely cover the bowl with aluminum foil. (DO NOT close hermetically, since air should pass over the fermenting milk). Leave it at room temperature (26 - 30° C), without touching it, for 24 hours. In order to obtain a good compound #2, during this time it is necessary to place the bowl on a working surface without vibrations and far from sources of heat. Remember to include today's date and time on the label.

END of DAY ONE
DAY TWO

Mix compound #1 with colostrum

1. Retrieve **compound #1**, which is in the yogurt maker's jars, which themselves are in the refrigerator. Using the tablespoon, stir it gently while inside the yogurt maker’s jars and then transfer 700 ml **(for one person 500ml)** of **compound #1** from the yogurt maker's jars into the measuring cup. Transfer this volume of **compound #1** to the big jar having one liter and half capacity.

Pour one bottle of bovine colostrum (125 ml) in the big jar having one liter and half capacity where you had just poured half 700 **(for one person 500)** ml of compound #1.

*Please notice, if you use certified raw colostrum, pour half a liter (500 ml) of bovine colostrum in the big jar having one liter and half capacity where you had just poured half a liter (500 ml) of compound #1.*

2. Stir very well this mixture of colostrum and compound #1 with the long tablespoon and leave the jar at room temperature for at least 12 hours, but not more than 24 hours. Cover the jar with aluminum wrap (do not close it with its own lid since air should enter).

3. **DO NOT DISCARD** the remaining amount of **compound #1**; keep it (in the glass cups), in the refrigerator. You may wish to include today's date on the label as well, in order to later confirm its expiration date.

*This surplus of **compound #1** will be re-used on **PROPAGATION** (see below) for the preparation of your next batch of MAF 314®.*
However, compound #1 cannot remain in the refrigerator for more than five days. If it is not used within five days, it has to be re-propagated anyway.
Final preparation of compound #2

1. In order to complete preparation of **compound #2**, after 24 hours in the glass bowl at room temperature, check that it is “thick” as expected and as shown in the course.

2. Stir it well with the table spoon to obtain a smooth and homogeneous compound. Then, filter the product through the strainer (to facilitate the filtration, you may use the table spoon or the ladle to help break up any remaining lumps through the strainer and into the one-liter jar).

3. Close the one liter jar with its lid without the rubber stopper, identify it as “**compound #2**” with a label, and place it into the refrigerator. You may wish to include today’s date on the label as well, in order to later confirm its expiration date.

You have now obtained the MAF 314® **compound #2**, now in the one liter jar which is itself in the refrigerator

**END of DAY TWO**
DAY THREE

1. Use the tablespoon to transfer 700 (for one person 500) ml of compound #2, previously prepared and placed into the one-liter jar now in the refrigerator, into the measuring cup.

   Please notice, if you use certified raw colostrum, use half a liter (500 ml) of compound #2.

2. Add this amount of compound #2 to the mixture - of colostrum and compound #1 - into the one-and-a-half liter jar. The one-and-a-half liter jar should therefore now be full. Stir it well with the long steel tablespoon.

   DO NOT DISCARD the remaining half-quart of compound #2; keep it in the one-liter jar, in the refrigerator. The surplus of compound #2 will be re-used on PROPAGATION for the preparation of your next batch of MAF 314®.

   However, compound #2 cannot remain in the refrigerator for more than five days. If it is not used within five days, it has to be re-propagated anyway.

4. Split the mixture (colostrum + compounds #1 and #2) from the big jar into 14 little glass jars 100ml each (7 little glass jars, 125 each, for one person) To do this, pour the mixture in the measuring cup and then transfer it in the little jars. Please be sure to record the date of preparation, so that you do not exceed the expiration date. You can use adhesive labels or other means.

   Important notice: if no product has gone lost, you have enough mixture (one liter and half) (for one person it is one liter plus 125ml of colostrums) to fill 14 little jars of 100 ml (7 little jars of 125ml for one person) capacity. This happens because the entire procedure has been designed to yield an excess of product, in order to provide a comfortable margin of safety in case mishaps or spills happen (or in case of sharing with another person). However, DO NOT EXCEED the recommended dose of MAF 314® in consumption according to
your physician’s indications. Further, remember that the expiration date for MAF 314® is no later than 8 days after preparation.

4. Finally, use the steel measuring spoon to add to each 100 ml jar (125 ml for one person) a smidgen of the MAF 314® probiotic #3. DO NOT MIX. Simply leave probiotic #3 on the top of each jar like a garnish.

5. Close the 100 ml jars (120ml jars for one person) securely, and place them into the refrigerator for at least 2 hours before consuming.

You have now produced MAF 314®, which is refrigerating in the small jars

END of DAY THREE
To consume MAF 314® safely as directed, please read the following instructions carefully.

**Suggested use:** one jar daily at the end of a meal rich in fibers (vegetables or leafy greens).

May be flavored adding high grade honey. **Sugar substitutes, sweeteners (contained also in light beverages, foods and chewing gums) deteriorate the product’s properties.**

Shake well before consuming.

**MAF 314® is a dairy product:** conserve refrigerated for no more than 8 days. **DO NOT FREEZE.**

**MAF 314® EXPIRATION DATE:** 8 days after preparation.

**MAF 314® is naturally formulated:** no additives, preservatives, artificial flavoring or coloring. It does not contain added sugar, gluten, derivatives of peanuts or nuts. Color, density and flavor may vary from batch to batch and over time.

**Ingredients:** cow milk and milk derivatives; bacteria; milk enzymes; yeasts.

**Calories** per 100 ml: 65 kcal.
- When you notice that only two daily doses of MAF 314® are left in the refrigerator, it’s time to begin preparing again the two compounds (compounds #1 and #2) to obtain your next batch of MAF 314®. In fact, since it takes 48 hours to prepare MAF 314®, if you start preparation when you still have two daily doses, you will have the next batch ready by the day after you have finished consuming the last dose.

The procedures described below are called PROPAGATION.

The propagation is very easy if you consider that

1 bag of powder of starter = six tablespoons

Don’t open a new bag of starter #1 and starter #2 every week. A new bag of starter #1 and of starter #2 should be opened every two months.

Instead of using a new bag of starter you must use six tablespoons of compound #1 and six tablespoons of compound #2 sitting in your fridge from the previous preparation.
PROPAGATION

Preparation of compound #1

Preparation of compound #1 is identical to that described in DAY ONE except for the fact that you do not open another bag of starter #1 but you add 6 (six) tablespoons of the compound #1 remained undisturbed into the fridge from the previous batch to one liter of milk (boiled and cooled down to 40-42°C).

Preparation of compound #2

Preparation of compound #2 is identical to that described in DAY ONE except for the fact that you do not open another bag of starter #2 but you add 6 (six) tablespoons of the previous compound #2 remained undisturbed into the fridge from the previous batch to one liter of milk (boiled and cooled down to 26-30°C).

PLEASE NOTICE: PROPAGATION of compound #1 and #2 may be repeated for no more than 10 times.

That is, after about TWO MONTHS, you have to start all over from scratch using a new bag of starter #1 and a new bag of starter #2.

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References


