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Comparison of chemical properties and biological activity of Immune Basics, Wellmune WGP yeast β -1,3 1,6-glucan with Immudyne nutritional quality β -glucan

Samples of the two commercially available β -1,3 1,6-glucan nutritional supplement Wellmune WGP Lot# 011811, 250 mg of β -glucan per capsule and Immudyne NQ Lot# 062110, final processing, nutritional quality β -glucan, were analyzed in parallel to compare points of similarity. As indicated in Table I the proximate analyses of these samples are similar. However, the impurities in WGP# 011811 such as ash, protein, and fats can contribute to some inhibition of uptake in the alimentary canal as well as binding to active sites of dendritic and macrophage cells needed to activate the immune processes.

Table I
Proximate Analyses

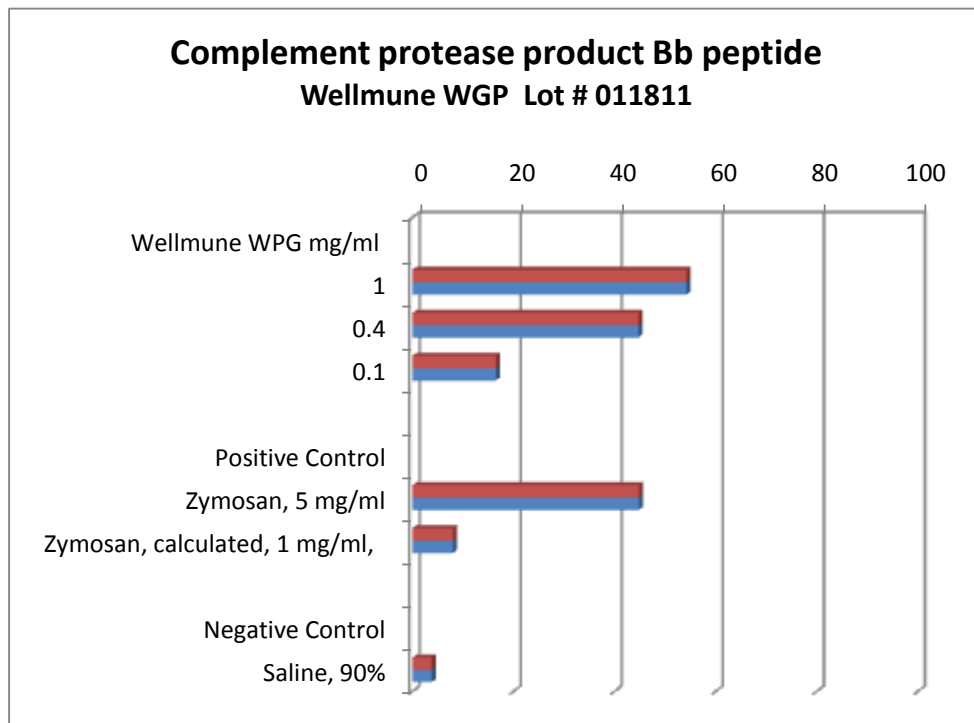
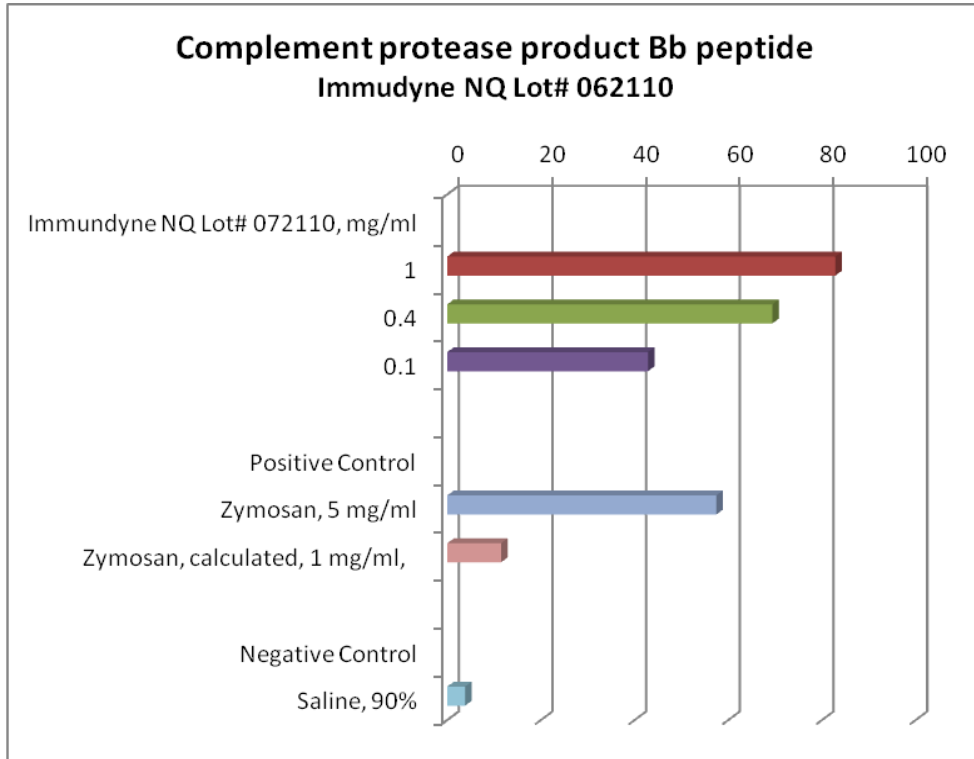
Compositional Analyses	Samples	
	Wellmune WGP Lot# 011811	Immudyne NQ Lot# 062110
Ash % by weight	1.63	<0.4
Kjeldahl nitrogen %, as ammonia	0.46	0.29
Percent protein % N x 6.25	2.88	1.81
Fats by extraction % wt	0.82	Not detected
Total Aerobic Plate Count	<10 cfu/gm	<10 cfu/gm

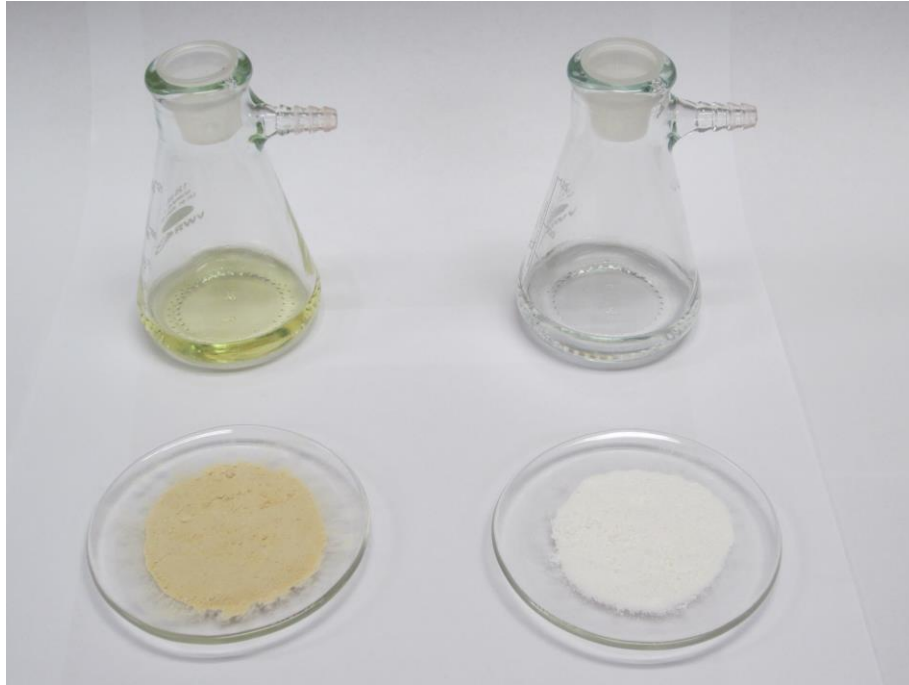
cfu = colony forming units

Table II
Complement Activation

Sample size mg/ml in serum tested	Complement protease product Bb peptide *	
	Wellmune WGP Lot# 011811	Immudyne NQ Lot# 062110
1	54.23	82.83
0.4	44.75	69.40
0.1	16.52	42.85

The complement activation capability of these glucans depends on the availability of the glucan chain to bind to the respective proteins in the secondary immune system pathway. The Immudyne NQ# 062110 gives greater responses for protease activity at all three concentrations of glucan in serum. Of particular interest is that even at the lowest concentration of 0.1 mg/ml serum the Immudyne NQ activates more than twice as much peptide cleavage as the WGP.

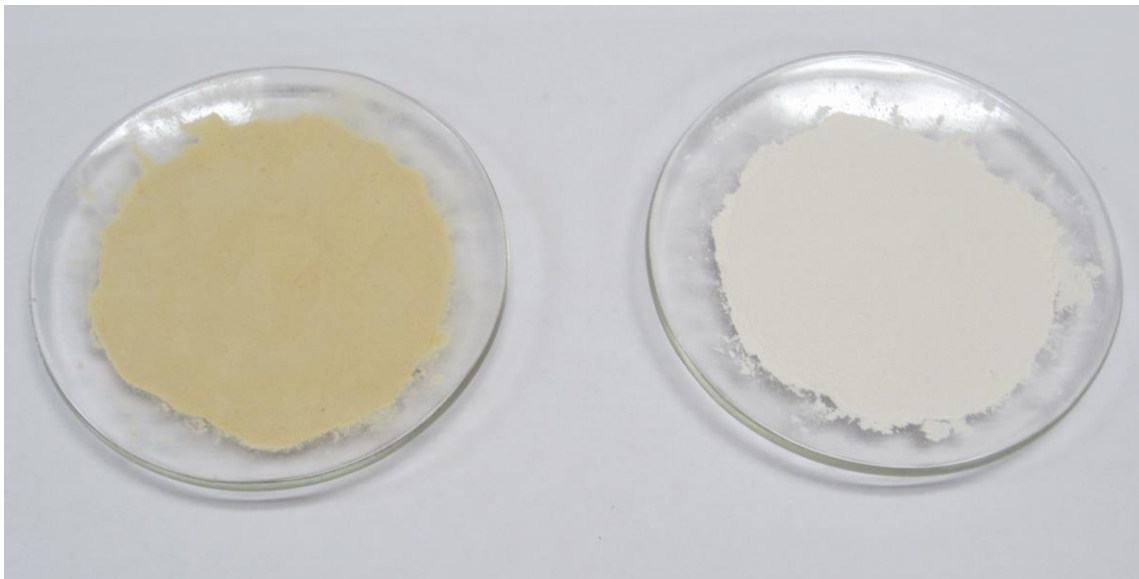




Wellmune WGP
Lot# 011811

Immudyne NQ
Lot# 062110

Extracts and Dried Powders



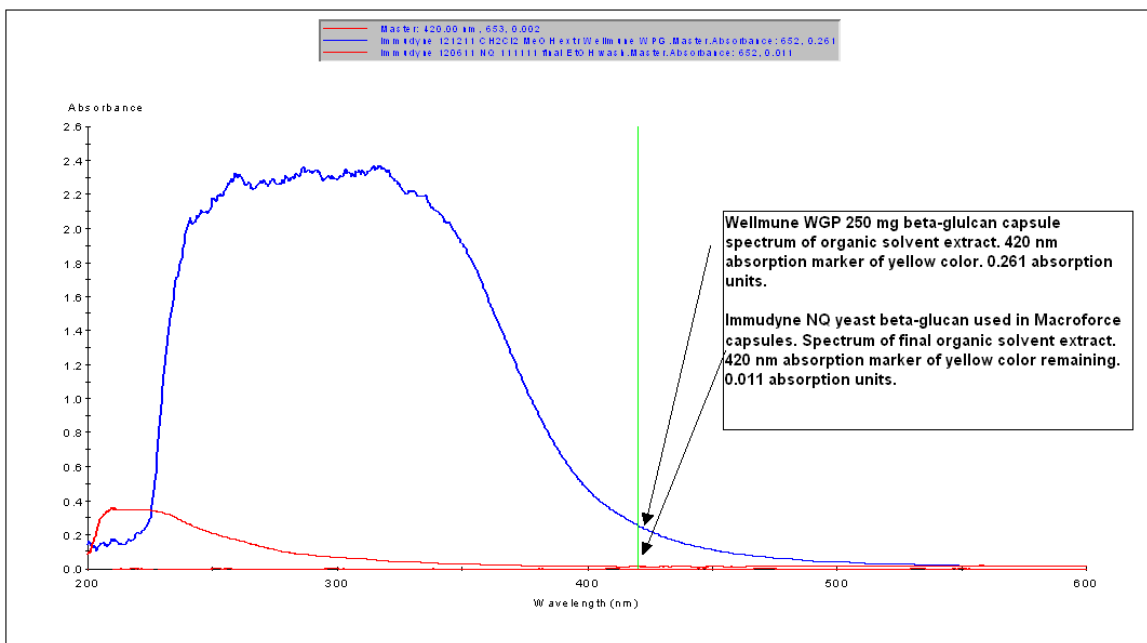
Wellmune WGP
Lot# 011811

Immudyne NQ
Lot# 062110

Unextracted Yeast β -1,3 1,6-glucan

Photography and ultraviolet-visible spectra of extractable impurities in the WGP and NQ samples

In the top photograph, labeled **Extracts and Dried Powders**, samples of Wellmune WGP and Immudyne NQ were extracted with organic solvent at room temperature to determine the relative quantity of pigmented orange fatty polymer still present in the nutraceuticals grade yeast β -glucans. The orange-brown powder from Wellmune WGP on the left yielded the orange solution above the powder in the photograph. The powdered samples were extracted and spectra determined on the extract after filtration. The extracted powders were dried of solvent for the recorded photograph. The absorbance of the extract at 420 nm is indicative of yellow to beige colorant in the extract. Although the extractions were not exhaustive the amount of color in the WGP is much greater than in the Immudyne NQ. According to the intensity of ultraviolet-visible absorbance at 420 nm the relative amount of yellow polymer being extracted from the WGP Lot# 011811 β -glucan is 24 times greater than in the Immudyne NQ Lot# 062110 according to the ultraviolet-visible spectrum below. In the lower photograph above, labeled **Unextracted Yeast β -1,3 1,6-glucan**, neither sample has had any of its colorant removed by further extraction. The Immudyne NQ product is as provided for the Macroforce capsules on the right of the picture and is obviously much more purified from the orange-brown fatty polymer that is a routine by-product of the industrial yeast cell wall glucan isolations.



Ultraviolet-visible spectra of extractable impurities in WGP and NQ samples

ASSIGNMENTS FOR CARBON-13				
NUCLEAR MAGNETIC RESONANCE SPECTRA				
December 13, 2011	File spectrum comparison		File spectrum comparison	
	B-1,3-Glucan		Wellmune WGP	
	Nayad '491		yeast B-glucan	
	Lot #052 - 01-07-91		Lot #011811	
Assignment	Standard ppm	Intensity height (mm)	250mg/capsule ppm	Intensity height (mm)
Aromatics from protein	None		none	
C-1, beta	103.7	111.5	102.9	84
C-1, beta	102.9			
C-1, beta				
C-1 alpha	None		None	
C-1 alpha				
C-1 alpha				
C-3	86.2	115.0	86.2	84
C-5	76.2	7.2	76.3	96
C-5	75.4	159.4	75.4	31
C-2	73.7	141.6	73.54	12
	73.3			
	72.7		72.76	93.0
C-6' (branched)	69.9	8.2	70.0	29
C-4	68.3	127.6	68.4	112
C-6	60.8	157.0	60.8	116
	39.6	DMSO, d-6, solvent.intensity. ca. 1000		
Lipid aliphatics - methylenes, olefins, and methyl groups	None		34	11
			30.8	19.0
			28.64	55
			28.1	24
			26.3	19.0
			24.23	19
			21.7	25
			13.5	23
			%WGP B-glucan,%	77.10
			%SGM	None
			%unknown	22.9
Ratio of C-6 intensity/ C-6' intensity (Degree of branching)		19.2		4.0
Ratio of C-3 intensity/ C-6' intensity (Degree of branching)		14.1		2.90

ASSIGNMENTS FOR CARBON-13				
NUCLEAR MAGNETIC RESONANCE SPECTRA				
December 13, 2011	File spectrum comparison	File spectrum comparison		
	B-1,3-Glucan	Immudyne B-glucan		
	Nayad '491	NQ Lot #062110		
	Lot #052 - 01-07-91			
Assignment	Standard ppm	Intensity height (mm)	ppm	Intensity height (mm)
Aromatics from protein	None		none	
C-1, beta	103.7	111.5	102.9	65
C-1, beta	102.9			
C-1, beta				
C-1 alpha	None		None	
C-1 alpha				
C-1 alpha				
C-3	86.2	115.0	86.2	66
C-5	76.2	7.2	76.5	25
C-5	75.4	159.4	76.3	76
C-2	73.7	141.6	73.35	21
	73.3		72.78	73
	72.7			
C-6' (branched)	69.9	8.2	70.0	16
C-4	68.3	127.6	68.4	77
C-6	60.8	157.0	60.8	91
	39.6	DMSO, d-6, solvent.intensity. ca. 1000		
Lipid aliphatics - methylenes, olefins, and methyl groups	None			
			25.2	18
			18.2	26
			% NQ B-glucan, % SGM	92.06
				None
				7.9
Ratio of C-6 intensity/ C-6' intensity (Degree of branching)		19.2		6.0
Ratio of C-3 intensity/ C-6' intensity (Degree of branching)		14.1		4.1

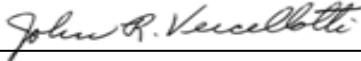
13-C nuclear magnetic resonance spectra of the WGP and NQ Samples

Both the WGP and NQ glucans qualitatively confirm the chemical shifts pertaining to all of the structural carbon atoms in the β -1,3 1,6-glucans under examination. The Immudyne NQ Lot# 062110 glucan has been isolated almost entirely free of nmr-detectable impurities. The only impurities found are small amounts of hydrocarbon resonances in the aliphatic region probably from trace amounts of fatty materials as well as solvent. The assignment of β -glucan purity to this NQ glucan is about 92%. The WGP Lot# 011811 has no other detectable impurities except a large amount of fatty hydrocarbon resonances. These could be part of the extractable fat in the proximate analysis above or most likely part of the highly colored fatty polymer carried through as extractable impurity and discussed above. The total β -glucan present is about 77% of the WGP solids. Neither the WGP nor the NQ contain detectable amounts of peptide linkage, aromatics, mannan, or phytyglycogen.

Conclusions concerning the relative quality of the WGP and Immudyne NQ β -glucans

The Immudyne NQ Lot# 062110 yeast β -glucan contains less detectable impurities by proximate analysis as well as by 13-C n.m.r. Both β -glucans have the essential backbone structure anticipated by their origins and isolation processes as demonstrated by their 13-C nmr spectra. The amount of organic solvent extractable impurity is much greater in the WGP glucan as demonstrated by ultraviolet-visible spectroscopy. Overall the quality of the yeast cell wall glucan in the product offered by Immudyne would be assumed to be favorable for macrophage activation as well as complement activation in the secondary immune pathway as the results of the serum protease complement activation indicated.

Approved by John R. Vercellotti, Ph.D.
Sharon V. Vercellotti Ms.C

Signature: 

Signature: 

Immudyne, Inc.