

Table S1. Forward and Reverse Primer Sequences

Gene	Forward Primer (5'→3')	Reverse Primer (5'→3')	Accession #
<i>ALP</i>	CTT GTG CCT GGA CGG ACC CT	TGG TGC ACC CCA AGA CCT GC	NM_000478.5
<i>NPP1</i>	CGC TGT TTC GAG AGA ACA TTT GGG A	AGT CGC CCT TGT CCT TGC AGT	NM_006208.2
<i>SOST</i>	CCA CCG GAG CTG GAG AAC AAC A	ATC GGT CAC GTA GCG GGT GAA G	NM_025237
<i>OPN</i>	TCA GCC AAA CGC CGA CCA AG	TGG AAG GGT CTG TGG GGC TAG GAG	AB469789.1
<i>GAPDH</i>	CCT GTT CGA CAG TCA GCC GCAT	TGG TGA CCA GGC GCC CAA TA	NM_002046.5

Table S2. Immunoblotting Protocol for Specific Proteins

Protein	Proteins of Interest (POI)				Housekeeping Proteins	
	ALP	NPP1	SOST	OPN	Vinculin	β -Actin
Protein Loaded	4 μ g	4 μ g	6 μ g	6 μ g	4 or 6 μ g (Depending on POI)	4 μ g
Stacking Gel	4%	4%	4%	4%	4%	4%
Running Gel	10%	8%	10%	10%	10%	8%
Sample Running	90 min @ 120 V	90 min @ 120 V	90 min @ 120 V	90 min @ 120 V	90 min @ 120 V	90 min @ 120 V
Membrane	0.2 μ m PVDF	0.2 μ m PVDF	0.2 μ m PVDF	0.2 μ m PVDF	0.2 μ m PVDF	0.2 μ m PVDF
Sample Transfer	60 min @ 100 V	60 min @ 100 V	60 min @ 100 V	60 min @ 100 V	60 min @ 100 V	60 min @ 100 V
Blocking	5% BSA/TBST for 1 h	5% BSA/TBST for 1 h	5% BSA/TBST for 1 h	5% milk/TBST for 1 h	5% BSA or milk (depending on POI)/TBST for 1 h	5% BSA/TBST for 1 h
1° Antibody	1:1000 rabbit (ab108337) in 5% BSA/TBST overnight	1:1000 rabbit (ab217368) in 5% BSA/TBST overnight	1:1000 rabbit (ab85799) in 5% BSA/TBST overnight	1:1000 rabbit (ab33046) in 5% milk/TBST overnight	1:1000 rabbit (ab129002) in 5% BSA or milk/TBST overnight	1:1000 rabbit (ab8227) in 5% BSA/TBST overnight
2° Antibody	1:10000 goat anti-rabbit (ab205718) in 5% BSA/TBST for 1 h	1:10000 goat anti-rabbit (ab205718) in 5% BSA/TBST for 1 h	1:10000 goat anti-rabbit (ab205718) in 5% BSA/TBST for 1 h	1:10000 goat anti-rabbit (ab205718) in 5% milk/TBST for 1 h	1:10000 goat anti-rabbit (ab205718) in 5% BSA or milk/TBST for 1 h	1:10000 goat anti-rabbit (ab205718) in 5% BSA/TBST for 1 h
ECL Substrate	Biorad Clarity™ Western for 5 min	Biorad Clarity™ Western for 5 min	Biorad Clarity™ Western for 5 min	Biorad Clarity™ Western for 5 min	Biorad Clarity™ Western for 5 min	Biorad Clarity™ Western for 5 min
Exposure	5 s	30 s	3 min	3 min	5 s	2 s