**Methods**

Study Area

The low budworm impact sites for this study were located in the Teanaway Community Forest in Washington State, approximately 40 miles from Central Washington University on public land. The study sites I analyzed were located near the following creeks: Stand Up Creek (903 m a.s.l), Jungle Creek (824 m a.s.l.), Jack Creek (963 m a.s.l.), and Moonbeam Creek (973 m a.s.l.). The high budworm impact sites were located in the Swauk drainage in the Okanogan-Wenatchee National Forest in Washington State, approximately 45 miles from Central Washington University and also on public land. These study sites were located near the following creeks: Cougar Creek (984 m a.s.l.), Hurley Creek (978 m a.s.l.), Hovey Creek (1050 m a.s.l.), and Blue Creek (1055 m a.s.l.).

The low and high budworm study areas are characterized by a mix of Douglas-fir (*Pseudotsuga menziesii)*, grand fir (*Abies grandis)*, ponderosa pine (*Pinus ponderosa*), western larch (*Larix occidentalis*) and at higher elevations, lodgepole pine (*Pinus contorta*). Summers (May-September) are relatively dry, with seasonal drought and temperatures ranging from 15°C-25°C, and winters (October-April) are wet with temperatures ranging from -5°C-11°C. The average precipitation for the area is 720 mm, mostly falling as winter snow (Northwest River Forecast Center, NOAA).

Budworm Life History

A major defoliator of the coniferous forests of Central Washington, as well as western North America in general (Senf et al. 2016), is the western spruce budworm (WSB) (*Choristoneura freemani)*—a native lepidopteran that ranges from southern British Columbia to Arizona and New Mexico (Fellin and Dewey, 1982). These insects emerge during budburst around mid-May to feed on the new growth of short needle conifers, specifically Douglas-fir and grand fir, and a few other species such as X and Y (Fellin and Dewey, 1982). Feeding continues until late June or early July when they then pupate and emerge as adults, taking flight around mid- to late-July for oviposition. Larvae then emerge the following year in mid-May to repeat their life cycle.

In a more natural fire regime that maintained an open forest structure, WSB outbreaks would occur about once every decade. In recent years, thicker forests from fire suppression and increased drought stress from climate change has created conditions that encourage more frequent and wider spreading WSB outbreaks (Willis et al, 2008; Lovett et al, 2006). This change in herbivore behavior as a result of shifted forest structure has the potential to change forest ecosystem dynamics with implications for forest-stream connectivity. Furthermore, the cold weather that would have normally killed off overwintering WSB larvae in the past occurs less often in the warmer climate,. This allows them to stay out longer, causing more damage to plants more often than they otherwise would (Griffin and Turner, 2012). It has also been suggested that WSB outbreaks can lead to increased fires due to the dead and dying trees left when an outbreak subsides (Schlesinger et al, 2015), but new research has shown that this may not be the case, and in fact may have the opposite effect.

Experimental Design

I used a nested study design with repeated sampling through time to investigate how budworm herbivory influenced throughfall composition, litter decomposition, and soil condition status. Within each budworm herbivory level (low versus high), I established study sites along 4 different streams (n=8). At each stream I established three replicate plots approximately 15 m from each other in an upstream–downstream direction. At each replicate plot, I measured throughfall water chemistry, litter decomposition rates, and soil chemistry 8 times between early September 2015 and early November 2016. Frass collectors were also deployed at each site to measure budworm activity as frass production over the course of the study. Throughfall and frass collectors were taken down during winter (date taken down – date put back up) to avoid damage by heavy snowpack.

Throughfall

At each plot (n= 24), a throughfall collector was installed under the canopy of a randomly selected tree. Each throughfall collector consisted of a funnel ( \_ mm diameter) that drained through tygon tubing into a 4-L acid-washed collection jug. To protect the tubing, I fed it through a PVC pipe, pounded into the ground with hole in the side so the tubing could enter the collection jug. The PVC pipe was stabilized by wiring it to a piece of rebar pounded into the ground. To prevent material from entering the collection jug, the opening was sealed with parafilm to keep the tubing in place, and polywool at the base of filter prevented litter from entering the jug from the funnel.

Upon rainfall, water entered the funnel and traveled through the tubing into jug until I retrieved within 48 h of the rain stopping. Upon collection, the total sample volume was recorded as the sample was transferred to an acid washed nalgene bottle and returned to the lab for filtration using a 1.0 μm glass fiber filter. Samples were frozen until later water chemistry analysis.

In order to differentiate nutrients in bulk rainfall compared to throughfall that had percolated through the canopy, a total of four rainfall collectors were set up in areas with no canopy cover, two in the low budworm study sites and two in the high budworm study sites.

Frass and Litter Measurements

To ensure a qualitative difference between low and high budworm herbivory, frass was collected at each site. Funnels of 0.25 m2 diameter made of tarp and garden hose connected to a one L Nalgene bottle were set up under trees at each site. These were sampled regularly during budworm feeding and less periodically after feeding and until snowpack. Unfortunately, because of frequent rains during feeding in 2016, no data is available due to samples decomposing before they could be collected and measured.

Litter decomposition

At each replicate plot I deployed twenty 20x20cm mesh litter bags (García-Palacios et al. 2016). Ten of those bags contained an air dried mixed conifer needle sample of Douglas-fir, grand fir, and ponderosa pine, the three most abundant species in the study area, in a mix of ~1.2 grams of ponderosa pine and then ~4.8 grams of mixed Douglas-fir and grand fir. The other ten bags at each replicate plot contained sugar maple (*Acer saccharum*) leaves which are non-native to the area but are commonly used in decomposition studies for comparison across biomes. I deployed a total of 480 bags across all sites.

Within each litter bag, I measured ~3-5 grams per bag of air dried leaves (Benfield, 1996), recorded initial leaf mass, and placed an aluminum tag with a unique ID. The bottom side of the mesh bags were made of a smaller sieve size (0.5 mm) (Schweitzer et al, 2005) than the top (2 mm) (Genung et al, 2013) to reduce the loss of its contents while still allowing small detritivores to enter the bags. Bags were assembled by stapling the two sieve sizes together and reinforcing them with super glue at the corners. The bags stayed intact throughout the 9 month deployment. Mesh bags with leaves were subsequently placed into bags with a larger mesh size (XX) for ease of handling and to further protect them during deployment, and each individual bag was placed into Ziplocs before deployment to capture transport and handing loss to correct initial mass.

In the field, bags were strung together on an approximately 6 m line held in place by a 2 ft piece of rebar driven into the ground on either side. The rebar anchors prevented bags from being moved by the wind or displaced by hillslope runoff. A coin flip determined which bags (conifers or deciduous maple) were placed upstream and downstream at each site. Handling loss was applied to the mass of the material by deploying twenty bags, ten deciduous and ten coniferous and extracting them immediately to see if there was any mass loss from deployment and extraction. This was done separately for conifer and deciduous leaves.

Bags were collected 7 times in approximately 1-2 month intervals with a 5 month break during winter snowpack when the sites were inaccessible. One conifer bag and one maple bag were cut and bagged in a Ziploc (to prevent mass loss) per site for a total of 48 bags per sampling time. On each retrieval from the field, bags were randomly chosen for pickup and returned to the lab in a Ziploc bag to prevent additional leaf mass loss. Upon return to the lab, decomposition bags were air dried in paper bags (Genung et al. 2013) on a clothesline to constant mass (Schweitzer, 2005). Once the bags were air dried, each bag was sorted to remove any noticeable debris that had become incorporated in the sample (Chapman et al. 2013). Because of natural loss of conifer needles from the canopy, it was difficult to determine what was originally in the bag and what had fallen into it, so the mass of conifer needles accumulated in the maple decomposition bags was sorted and used as a correction factor for the mass of conifer needles that entered the conifer bags.

*Soil Analyses*

Upon each collection of decomposition bags, I also used a thermocouple to measure temperature of the O horizon at a depth of ~2 cm, at the top of the A horizon at a depth of 10 cms and at 20 cm depth in the A horizon. A soil core of ~10 cm depth was also collected from each replicate plot at each stream site each time I collected litter bags. Soil cores were stored on ice for return to the laboratory whereupon each core was homogenized in a Ziploc bag. Soils were analyzed for moisture content, percent organic matter, ammonia, nitrate, organic N, inorganic P, and organic P using methods detailed below.

*Moisture Content and Percent Organic Matter:*

Soil was sieved at 2 mm and a subsample was placed into an ashed aluminum pan and weighed immediately for field mass. Pans were then placed in a drying oven at 60ºC until constant mass, cooled to room temperature, and weighed to obtain dry mass (DM). The difference between field mass and dry mass was used to calculate percent moisture. Then dried soil samples were placed in a muffle furnace at 500ºC for 48 hrs. to combust all organic compounds. After ashing, samples were cooled to room temperature, rehydrated with Milli-Q water to rehydrate and colloids containing water molecules, and then placed again into a drying oven until constant mass. Pans were cooled to room temperature and reweighed, with the difference between dry mass and ash free dry mass used to calculate percent organic matter.

*Phosphorous Analysis*

The Bray P1 method was used to extract phosphorus from each soil sample (Hamilton, 1997; Patton and Kryskalla, 2003). To do this, one gram of air dried soil was added to 10 mLs of the Bray P1 extractant solution (30 mLs 1 N NH4F to 50 mLS 0.5 HCl) and was shaken on a shaking table at 100 rpm for 15 minutes. The sample was then filtered with a syringe through a 0.5 µm glass fiber filter and stored in the freezer until analysis. Samples were analyzed for inorganic phosphorous on a Seal AQ1 Discrete Analyzer (Seal AQ1, Seal Analytical; Mequon, Wisconsin, USA) with EPA equivalent methods.

*Nitrogen Analyses*

A 2M KCl extraction method was used to extract inorganic nitrogen from each soil sample. Ten grams of air dried soil were added to 70 mLs of 2M KCl and shaken at 100 rpm for 2 hours on a shaker table. The sample was then filtered with a syringe through a 0.5 µm fiberglass filter and stored in the freezer until analysis. Samples were analyzed for NO3- using cadmium reduction and NH4+ phenate method on a Seal AQ1 Discrete Analyzer ((Seal AQ1, Seal Analytical; Mequon, Wisconsin, USA) using EPA equivalent methods.

Each site also contained a resin bag made of bleached nylons (to prevent color leaching that may affect results) filled with 30 grams of ion exchange beads to measure the activity in soils and was also extracted using 2M KCl and were analyzed for NO3- and NH4+ to look at net nitrification on a Seal AQ1 Discrete Analyzer using EPA equivalent methods. These bags were deployed 10 cms deep in the soil during while initial soil samples were taken to examine the change in soil chemistry just before snowpack and just after snow melt.

**Statistical Analysis**

I used a two-sample t-test to compare the two treatments; coniferous litter vs deciduous. This was done for both the rate of decomposition and for net nitrification. I used generalized least squares (GLS) models and linear mixed effects (LME) models (Senf et al. 2016) to compare the variances within each of the 8 sites and each of the 3 replications within to compare percent soil moisture, percent organic matter, NO3-, NH4+, SRP, N:P ratio, total inorganic N, and net nitrification/mineralization. Data was analyzed using R.