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A Bootstrapping Approach for Investigating the Consistency of Assignment of Plants to Jamu Efficacy by PLS-DA Model

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ABSTRACT

Present study investigates the main ingredient plants in Jamu medicines using PLS-DA where the model was developed by considering plants usage in Jamu as predictors and Jamu efficacy as response. We utilized the coefficient matrix obtained from the PLS-DA model to assign plants to Jamu efficacy based on the largest coefficients. However, if new Jamu data set is added to the model, the coefficient configuration, and in turn the assignments, may change. Thus, consistency examination of the assignments is important and bootstrapping can be used for this purpose. If a plant is useful for certain efficacy then in most Bootstrap resampling rounds the plant would be assigned to that efficacy and most of the plant's coefficients corresponding to that efficacy are expected to be positive. In the present study 1000 Bootstrap rounds were performed. Out of 465 plants, it is found that the assignments of 276 plants are consistent and these plants are regarded as main ingredient in corresponding Jamu. Thus, this study gives useful information on plants serve as main ingredients in Jamu efficacy by evaluating the significance of plant usage in Jamu medicines using Bootstrap procedure.

Keywords: Jamu, PLS-DA, Bootstrap.

1. INTRODUCTION

Jamu is the common name for Indonesian herbal medicines. The ingredients, i.e. plants composition, play important role in determining the Jamu efficacy. Among the ingredients of Jamu are plants used as main ingredients, which contribute primarily to its efficacy, as well as plants used as supporting ingredients (Pramono (2007)). Investigating which plants are main ingredients and which are supporting is important to comprehensively understand the mechanism of plants used in Jamu to achieve specific efficacies.

A Partial Least Square Discriminant Analysis (PLS-DA) model can be helpful in this attempt by relating plants usage in Jamu as predictors and Jamu efficacy as response. PLS-DA is suitable for this analysis regarding that large number of plants are used in Jamu and on the other hand, Jamu efficacy are in categorical scale Plants perform as main ingredients will have significant effect on the model developed.

However, PLS-DA was designed mainly for prediction of the responses by maximizing the relationship between predictors and responses (Wold *et al.* (2001); Marker and Rayens (2003) and Boulesteix and Strimmer (2006)). PLS-DA gives little attention on the effect of each predictor on responses as it did not provide significance testing on the coefficient. So, if new Jamu data set becomes available and added to the model, the coefficient configuration and in turn the assignments, may change. Thus, consistency examination on this assignment is important. To solve this problem, we conducted Bootstrapping, which is one of the methods that provide variance of a statistic whenever the variance of the statistic has no closed form or whenever the assumption of the theoretical distribution for the statistic is not satisfactory (Efron and Tibshirani (1993) and Manly (1997)).

The Bootstrap works by resampling the data set many times, calculates the same statistic in each resampling round, and accumulates the statistic value obtained in each round to become the distribution for the statistic. Variance of this distribution serves as variance of the statistic; whereas the distribution can also be used as empirical distribution of the statistic (Efron and Tibshirani (1993) and Manly (1997)). This Bootstrap procedure is suitable as a basis for consistency examination of the PLS-DA coefficient based assignment. The resampling step can be viewed as an attempt to generate new Jamu data set; whereas the assignment on each resampling round and its accumulation over all rounds can be used to check the assignment stability. In addition, accumulation over all resampling rounds of PLS-DA coefficients obtained on each round provides distribution of the coefficient. If a given plant is useful for certain efficacy then in most Bootstrap resampling rounds the plant would be assigned to the efficacy and most of the plant’s coefficients on that efficacy are likely to be positive. Thus, this study gives useful information on plants serve as main ingredients in Jamu efficacy by evaluating the significance of plant usage in Jamu medicines using Bootstrap procedure.

2. MATERIALS AND METHODS

a) Data set

In the present study, the commercial Jamu registered at The National Agency for Drug and Food Control (NA-DFC) of Indonesia are used in PLS-DA modeling. The data contain 3138 Jamu and in total they use 465 plants. Each Jamu is classified into one of nine efficacy categories, namely: (1) disorders of appetite (DOA), (2) disorders of mood and behavior (DMB), (3) female reproductive organ problems (FML), (4) gastrointestinal disorders (GST), (5) musculoskeletal and connective tissue disorders (MSC), (6) pain/inflammation (PIN), (7) respiratory disease (RSP), (8) urinary related problems (URI), and (9) wounds and skin infections (WND). All data used in this analysis can be accessed at http://kanaya.naist.jp/jamu/top.jsp.

b) PLS-DA modeling

Let n denotes the number of Jamu (in this case, 3138) and m denotes the number of plants (in this case, 465) used by these n Jamu. The usage status of plant i on Jamu $h(x_{hi}; h=1,2,\ldots,n; i=1,2,\ldots,m)$, which is equal to 1 if plant i is used in Jamu h and equal to 0 otherwise, serves as the predictors in PLS-DA; whereas the efficacy status of Jamu h on efficacy $j(y_{hi}; j=1,2,\ldots,9)$, which is equal to 1 if Jamu h is classified into efficacy j and equal to 0 otherwise, provides the responses in PLS-DA.

The details of the PLS-DA modeling are as follows (Marker and Rayens (2006) and Wold *et al.* (2003)). Let $T(n\times k)$ is a matrix of the underlying factors of X and is obtained by maximizing its covariance with the corresponding matrix of the underlying factors of Y, that is

$$T = XW \tag{1}$$

where $W(m\times k)$ is a matrix of weight and k is the number of factors extracted. Matrix T, multiplied by matrix of X –loadings $P(m\times k)$, is a good summaries of X

$$X = TP^t + E \tag{2}$$

so that the X –residuals $E(n \times m)$ is small. In addition, matrix T is also a good predictor of Y

$$Y = TQ^t + F \tag{3}$$

where $Q(9 \times k)$ is matrix of Y –loadings. The Y –residuals $F(n \times 9)$ express the deviation between the observed and the predicted responses.

Substituting Equation (1) into Equation (3) we obtain multiple regression model of PLS-DA

$$\mathrm{Y} = \mathrm{XWQ}^{\mathrm{t}} + \mathrm{F} \tag{4}$$

where the response prediction can be calculated as

$$\widehat{\mathrm{Y}} = \mathrm{XWQ}^{\mathrm{t}} = \mathrm{XC} \tag{5}$$

and the PLS-DA coefficient matrix $C(m \times 9)$ is calculated as

$$\mathrm{C} = \mathrm{WQ}^{\mathrm{t}} \tag{6}$$

c) Assignment of plants to the efficacy of Jamu

The efficacy of Jamu is determined by the plants used as its ingredients. Note that a plant can be used by many Jamu that have different efficacies. On the other hand, the matrix of PLS-DA coefficient C contains information on the effect of predictors on responses, i.e. the effect of plants on the efficacy of Jamu. Therefore, the matrix C can be utilized to assign plants to the efficacy for which they are most useful. The method of this assignment is as follows.

Due to the binary nature of y_{hj}, which contains information on the efficacy of Jamu h (i.e. $y_{hj} = 1$ if Jamu h is useful for efficacy j and $y_{hj} = 0$ otherwise), a large value of $\hat{y}_{hj}$ will lead to the prediction that Jamu h is useful for efficacy j. On the other hand, a given plant with a positive or negative coefficient in matrix C contributes positively or negatively to the value of $\hat{y}_{hj}$, respectively.

Considering these, a plant i is assigned as useful for efficacy j if its coefficient on efficacy j is positive. Let C_{ij} be a coefficient of plant i on efficacy j and U_{ij} be an assignment status of plant i on efficacy j. Thus,

$$U_{ij} = \begin{cases} 1; & \text{if } C_{ij} > 0 \\ 0; & \text{otherwise} \end{cases}$$

Furthermore, if plant i is predicted to be useful for efficacy j then this plant should be used by some Jamu having efficacy j. We verify our prediction as follows. Let W_{ij} be the number of Jamu with efficacy j that use plant i and is calculated as

$$W_{ij} = \sum_{h=1}^{n} X_{hi} Y_{hj}.$$

If $U_{ij} = 1$ and $W_{ij} > 0$ then the assignment of plant i as useful for efficacy j is called as Hit on the contrary, if $U_{ij} = 1$ and $W_{ij} = 0$ then the assignment is called as Miss.

d) Bootstrapping PLS-DA model

The concept of the Bootstrap used in the present study is shown in Figure 1. The details are as follows.

Step 1. Resampling of Jamu data.

This step is intended to generate new Jamu data set by resampling the existing Jamu data set. As for other resampling method for the Bootstrap, the resampling is performed with replacement with sample size equal to the original data set, i.e. 3138. Due to the replacement process during resampling, some Jamu may be not selected, selected only once or selected more than once although the total number of Jamu after the resampling is equal to the original data set. Thus, the Jamu configuration among the original data after resampling will be different.

Step 2. PLS-DA modeling in resampling data set.

Let n denotes the number of Jamu in the new Jamu data set obtained in Step 1, which is equal to 3138 and m_b is the number of plants used in n Jamu. Note that m_b may be less than the total number of plants in the original data set m because some plants may not be included in m_b if Jamu that use the plants are not selected during resampling in Step 1. Following previous definition, let $\tilde{x}_{hi}(h=1,2,\ldots,n;\ i=1,2,\ldots,m_b)$ is the usage status of plant i on Jamu h, which is equal to 1 if plant i is used in Jamu h and equal to 0 otherwise; whereas $\tilde{y}_{hj}(j=1,2,\ldots,9)$ is the efficacy status of Jamu h on efficacy j, which is equal to 1 if Jamu h is classified into efficacy j and equal to 0 otherwise.

Using this definition, PLS-DA model is performed, where the matrix $\widetilde{\mathbf{X}}$ and $\widetilde{\mathbf{Y}}$ provide the predictors and responses, respectively. The coefficient matrix obtained is denoted by $\widetilde{\mathbf{C}}$ $(m_b \times 9)$, whereas A_{ijr} is the assignment status of plant i on efficacy j in Bootstrap round r, which is equal to 1 if in Bootstrap round r plant i is assigned to efficacy j and equal to 0 otherwise.

Step 3. Accumulation of PLS-DA coefficients and assignment results.

The PLS-DA coefficients and the assignment results from Step 2 are accumulated into coefficient distribution and Bootstrap assignment probability, respectively. Let R_i is the number of plant i selected in R Bootstrap rounds and B_{ij} is the coefficient distribution of plant i on efficacy j, which contains R_i coefficient values. The Bootstrap assignment probability of plant i on efficacy j, T_{ij} is calculated as

$$T_{ij} = \sum_{r=1}^{R} A_{ijr} / R_i. \tag{7}$$

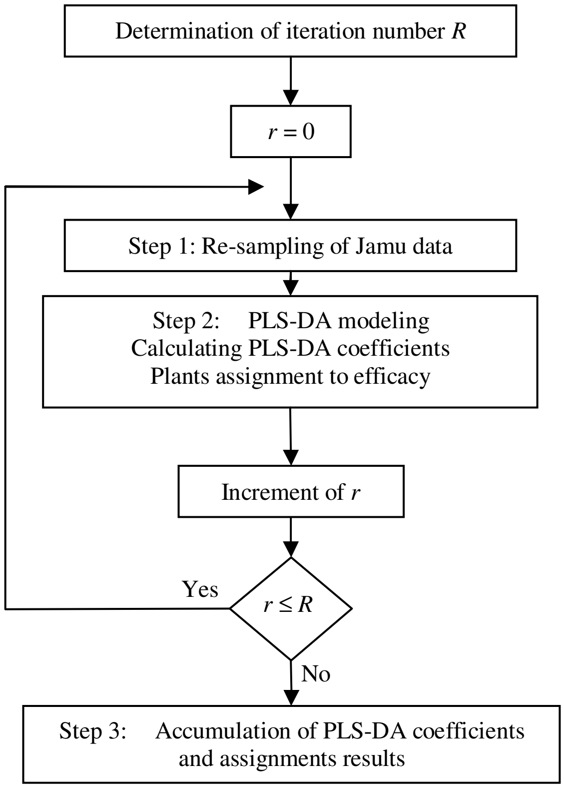


Figure 1: The schematic diagram of Bootstrap steps used in the present study

Following the procedure above, the goal of the Bootstrap used in the present study is to obtain coefficient distribution as well as Bootstrap assignment probability of plant i to efficacy j, B_{ij} and T_{ij} respectively. After that, these two statistics are then used in examining the assignment consistency of plant i to efficacy j. The idea in the examination is that if plant i is useful for efficacy j then plant i will be assigned frequently to efficacy j during the resampling process, i.e. the value of T_{ij} should be large enough. In addition, most coefficients corresponding to plant i on efficacy j, B_{ij} should be positive.

Combining these two criteria, assignment of plant i on efficacy j using PLS-DA model is said to be consistent if: (1) $T_{ij} \geq 0.8$ and (2) $P(B_{ij} < 0) \leq 0.05$. The second criterion is equivalent with Percentile 5% $P5 > 0$.

3. RESULTS AND DISCUSSION

In selecting the number of components for PLS-DA model, we performed 5-folds cross validation and calculated Prediction Error Sum of Square (PRESS) to measure the deviation between observed and predicted responses. It is observed that the PRESS statistic is the smallest at 10 components. Hence, we use 10 components in PLS-DA model for the original Jamu data set as well as for the Bootstrapping process.

By using positive coefficient as a criterion in assigning plants to a given efficacy, we obtain the Hit–Miss status, as summarized in Table 1, which indicates that there are many plants categorized as a Miss. In other words, these plants are assigned as useful for a certain efficacy because they have a positive coefficient on that efficacy, but no existing Jamu with that efficacy uses those particular plants.

TABLE 1: Assignment status of plants to efficacy

Efficacy	Assignment using positive coefficient		Assignment using maximum coefficient	
	Hit	**Miss**	**Hit**	**Miss**
URI	48	32	23	0
DOA	94	8	45	0
DMB	35	72	13	0
GST	149	8	82	1
FML	115	50	61	4
MSC	172	6	94	0
PIN	113	25	69	0
RSP	62	81	31	0
WND	86	11	42	0

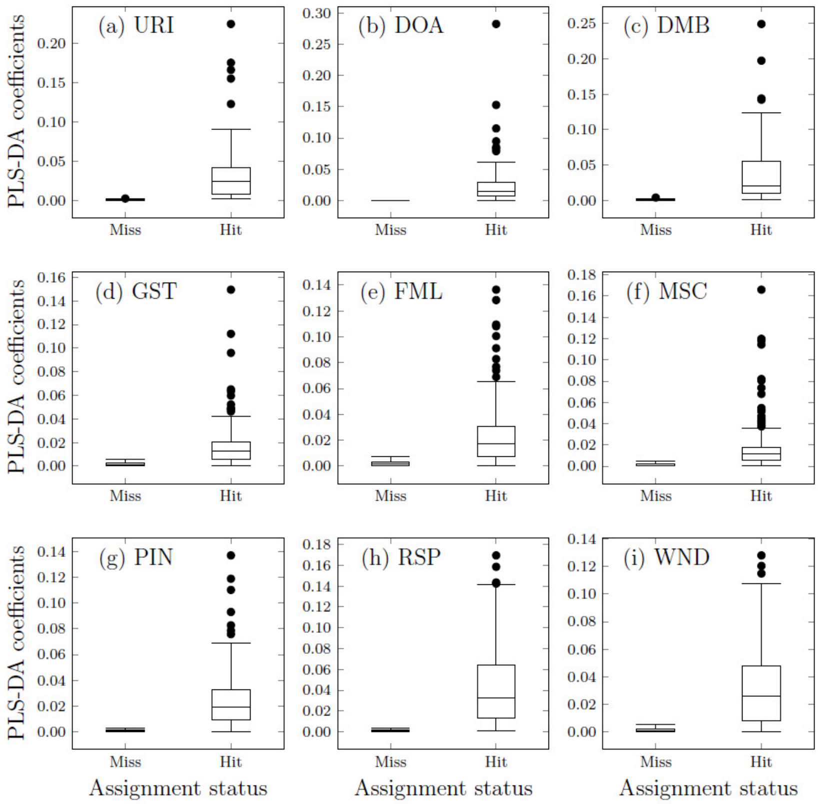


Figure 2: Distribution of the PLS-DA coefficients corresponding to the assignment status using positive coefficient.

Figure 2 shows the distribution of PLS-DA coefficients corresponding to the assignment status using positive coefficient. From Figure 2, it is obvious that plants categorized as Miss have coefficient values very near to zero. Therefore, it is evident that the quality of prediction of efficacies for plants can be improved by considering not only the sign (i.e. negative or positive) but also the magnitudes of the coefficients.

In order to reduce the number of Miss categorizations, we further improved our assignment of plants to specific efficacies, as follows. Note that each plant has nine coefficients, one for each efficacy. Rather than assigning plants to each efficacy where they have positive coefficient, the new assignment allocates a plant to an efficacy only when the plants in question has the largest coefficient for that efficacy. Thus, if A_{ij} denotes the new assignment status of plant i to efficacy j, then

$$A_{ij} = \begin{cases} 1; \text{ if } C_{ij} = \max_j (C_{ij}) \\ 0; \text{ otherwise} \end{cases}.$$

Note that on previous assignment, a given plant may be regarded as useful for more than one efficacy, i.e. $\sum_{j=1}^{9} U_{lj} \geq 1$, whereas for this new assignment a given plant is regarded as useful for one efficacy only, i.e. $\sum_{j=1}^{9} A_{lj} = 1$. For this new assignment, if $A_{lj} = 1$ and $W_{lj} > 0$ then the assignment of plant l regarded as useful for efficacy j is called as Hit, whereas if $A_{lj} = 1$ and $W_{lj} = 0$ then the assignment is called as Miss. Table 1 shows summary of Hit–Miss status based on this new method of assignment. The recognition rate for Hit–Miss status is 98.9%, which corresponds to the fact that the number of Miss categorizations for the new method is only five plants out of a total of 465.

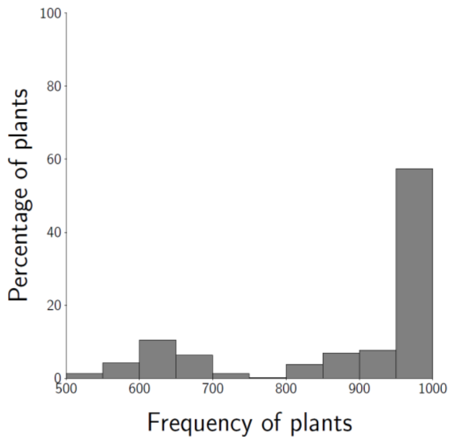


Figure 3: Distribution of frequency of plants selected in 1000 bootstrap rounds.

After the assignment is performed, then The Bootstrapping is conducted to examine the consistency of this assignment. The number of Bootstrap resampling rounds R is 1000 in this study. However, a given plant may be selected less than 1000 times because Jamu that use the plant might not be selected during some resampling rounds. Among 1000 bootstrap rounds, the least number of times a plant is selected is 504 and most of them are selected more than 950 times (Figure 3).

As indicated in Equation (7), the Bootstrap assignment probability, T_{ij}, indicates the proportion of plant i assigned to efficacy j over all resampling rounds. Figure 4 shows the plots of Bootstrap assignment probability against percentile 5% of the coefficient distribution P5 for all the plants corresponding to efficacies predicted for them using maximum coefficient method. In Figure 4 the range of T_{ij} varies from 0.182 to 1. Among all 465 plants, 291 plants (62.6%) have Bootstrap assignment probability greater than the threshold 0.8. No plant has Bootstrap assignment probability greater than the 0.8 corresponding to efficacies not predicted for them implying the effectiveness of the maximum coefficient method.

Meanwhile, if plant i is assigned to efficacy j then the coefficient C_{ij} itself should be significantly larger than 0 so that the assigned plant can be considered as significantly affecting the efficacy. This requirement can be checked by utilizing the coefficient distribution of plant i on efficacy j, B_{ij} obtained from the bootstrapping. As noted in the previous section, the percentile P5 of B_{ij} should be greater than 0, which indicating that the plant i is affecting the efficacy j at 5% significant level. Figure 4 shows the percentile P5 of the corresponding plants assigned using the maximum coefficient method. Over all 9 efficacies, we found 326 plants (70.1%) have positive P5 on the efficacies which were assigned to the plants using the maximum coefficient method.

Considering the correlation value ρ (Figure 4), the Bootstrap assignment probabilities are positively correlated with percentile P5. It means that the larger the Bootstrap assignment probability of a given plant the larger is its percentile P5. It is reasonable since the assignment is conducted based on maximum coefficient. The larger the coefficient of a given plant on an efficacy, the larger its percentile P5 and the probability is high that the assignment of the plant to that efficacy is correct.

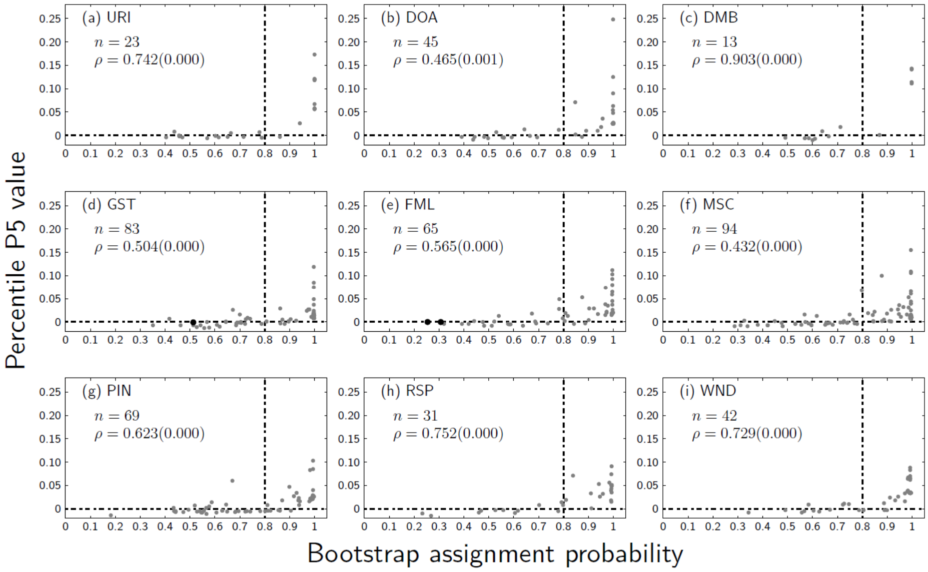
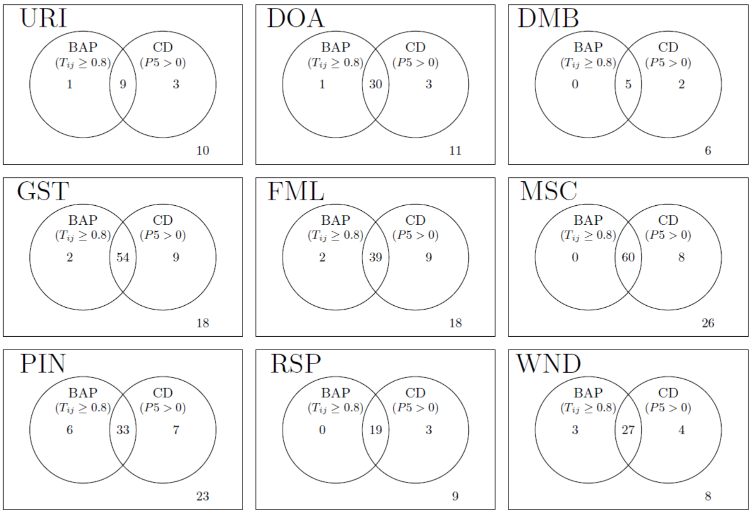


Figure 4: Plot of Bootstrap assignment probability versus percentile P5 value of plants assigned to the efficacy. This figure gives plot between the Bootstrap assignment probability and percentile P5 value of plants assigned on each of the nine efficacy groups. The dashed line is created to help identify plants fulfilling the two criteria of consistency. The black dots represent plants classified as Miss. The number of plants assigned to each efficacy is denoted as n. The correlation value between the two variables is denoted as ρ, where the figures in parentheses is its p-value.

The relation between Bootstrap assignment probability and percentile P5 value can be explored more by using the scatter plot of Figure 4. In the figure, two lines drawn at points 0.8 and 0 for Bootstrap assignment probability and percentile P5 value, respectively, are used to help distinguish plants satisfying two criteria of consistency, i.e. $T_{ij} \geq 0.8$ and P5 > 0. In total for all 9 efficacies, we found 276 plants satisfying both criteria, 65 plants satisfying only one criterion, and 124 plants not satisfying any of the two criteria. The detail of the number of such plants for each efficacy is shown in Figure 5. Thus, 276 plants satisfying both criteria of consistency are regarded as consistent and predicted to act as main ingredients in corresponding Jamu. Scientific literature supports that all of these 276 plants whose assignments are regarded as consistent are useful for the corresponding efficacy. Moreover, in Figure 4, plants regarded as Miss are represented as green points. Among these Miss plants, none of them are consistent. Thus, we can obtain major plants, i.e. plants as main ingredients, corresponding to individual efficacies, which are informative to propose formulas of new Jamu medicines.



Note: (1) BAP: Bootstrap assignment probability
(2) CD: Coefficient distribution

Figure 5: Venn diagram showing the number of plants that satisfy both criteria, one criterion, and neither of the two criteria.

4. CONCLUSIONS

This work investigates which plants act as main ingredients in Jamu using Partial Least Square Discriminant Analysis (PLS-DA) by relating plants usage in Jamu (as predictors) with Jamu efficacy (as response). From the PLS-DA model, we utilized the coefficient matrix to assign plants to Jamu efficacy based on the largest coefficients.

Bootstrap can provide a basis for examining the assignment consistency of plants to Jamu efficacy based on PLS-DA coefficient. An assignment involving a given plant to an efficacy is considered as consistent when in most Bootstrap resampling rounds the plant is assigned to the efficacy and most of the plant's coefficients on that efficacy accumulated over all Bootstrap rounds are positive. Among 465 plants, we found assignments of 276 plants as consistent and the usages of all of them on the corresponding efficacy are supported by scientific literature.

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Appendix A: List of plants with consistent assignment based on Bootstrap procedure

No	Plant	No	Plant
Efiicacy: urinary related problems (URI)			
1	Imperata cylindrica	6	Pygeum africanum
2	Prunus cerasus	7	Serenoa repens
3	Strobilanthes crispus	8	Sonchus arvensis
4	Orthosiphon stamineus	9	Paeonia suffruticosa
5	Cucurbita pepo		
Efficacy: disorders of appetite (DOA)			
1	Litsea chinensis	16	Coleus forskohli
2	Benincasa hispida	17	Commiphora wightii
3	Zingiber purpureum	18	Avena sativa
4	Cassiae obtusifolia	19	Jasminum pubescens
5	Polygonum multiflorum	20	Lonicera japonica
6	Garcinia cambogia	21	Albizzia falcataria
7	Jatropha curcas	22	Cassia angustifolia
8	Tectona grandis	23	Nelumbo nucifera
9	Guazuma ulmifolia	24	Crataegus pinnatifida
10	Carum carvi	25	Caralluma fimbriata
11	Rheum tanguticum	26	Theae sinensis
12	Murraya paniculata	27	Curcuma heyneana
13	Terminalia catappa	28	Curcuma soloensis
14	Cassia tora	29	Cassia fistula
15	Amorphophallus konjac	30	Ilex paraguariensis
Efficacy: disorders of mood and behavior (DMB)			
1	Eleutherococcus senticosus	4	Brassica nigrae
2	Ipomoea reptana	5	Valeriana javanica
3	Polygala glomerata		
Efficacy: gastrointestinal disorders (GST)			
1	Artemisia annua	28	Sechium edule
2	Clematis armandii	29	Litchi chinensis
3	Cynara scolimus	30	Ledebouriella divaricata
4	Angelica keiskei	31	Alpinia officinarum
5	Allium ursinum	32	Boswellia carteri
6	Vaccinium myrtillus	33	Phaleria papuana
7	Ribes nigrum	34	Swietenia mahagoni
8	Pandanus conoideus	35	Swietenia macrophylla
9	Croton tiglium	36	Morinda citrifolia
10	Chlorella vulgaris	37	Ananas comosus
11	Syzygium cumini	38	Momordica charantia
12	Tanacetum parthenium	39	Euphorbia thymifolia
13	Coptis chinensis	40	Musa balbisianna
14	Rubus rosaefolius	41	Rauvolvia serpentina
15	Gymnema sylvestre	42	Vernonia cinerea

No	Plant	No	Plant
16	Magnolia officinalis	43	Symplocos odoratissima
17	Psidium guajava	44	Schisandra chinensis
18	Citrus amblycarpa	45	Apium graveolens
19	Simmondsia chinensis	46	Silybum marianum
20	Archangelisia flava	47	Spirulina
21	Melaleuca leucadendra	48	Fragaria vesca
22	Cocos nucifera	49	Matricaria chamomilla
23	Canarium commune	50	Scaphium affinis
24	Lindera strychnifolia	51	Gynura pinnatifida
25	Brassica napus	52	Hibiscus mutabilis
26	Phoenix dactylifera	53	Ziziphus spina-christi
27	Brucea javanica	54	Daucus carota
Efficacy: female reproductive organ problems (FML)			
1	Garcinia atroviridis	21	Kaempferia angustifolia
2	Tamarindus indica	22	Curcuma longa
3	Allium fistulosum	23	Ocimum sanctum
4	Pluchea indica	24	Galla lusitania
5	Ficus benjamina	25	Quercus lusitanica
6	Cimicifuga racemosa	26	Aglaia odorata
7	Luffa cylindrica	27	Prunus persica
8	Curcuma phaeocaulis	28	Areca catechu
9	Erythrina variegata	29	Trifolium pratense
10	Erythrina hypaphorus	30	Piper betle
11	Punica granatum	31	Sparganium stoloniferum
12	Ligusticum acutilobum	32	Nyctanthes arbor-tritis
13	Lepidium meyenii	33	Artocarpus communis
14	Elaeocarpus ganitrus	34	Ficus deltoidea
15	Terminalia bellirica	35	Lantana camara
16	Phaseolus radiatus	36	Solanum verbacifolium
17	Sauropus androgynus	37	Pouzolzia zeylanica
18	Parameria laevigata	38	Tetranthera brawas
19	Mirabilis jalapa	39	Sesbania grandiflora
20	Coriandrum sativum		
Efficacy: musculoskeletal and connective tissue disorders (MSC)			
1	Arisaema consanguineum	31	Alpinia galanga
2	Asparagus officinalis	32	Zingiber zerumbet
3	Atractylodis Macrocephala	33	Zingiber aromaticum
4	Hordeum vulgare	34	Languas galanga
5	Allium tuberosum	35	Psoralea corylifolia
6	Pterospermum javanicum	36	Massoia aromatica
7	Piper retrofractum	37	Setaria italica
8	Capsicum frutescens	38	Melia azedarach
9	Erythroxylum catuaba	39	Ptychopetalum uncinatum
10	Cordyceps sinensis	40	Artocarpus heterophyllus
11	Ruellia tuberosa	41	Myristica argentea
12	Cola nitida	42	Eurycoma longifolia

No	Plant	No	Plant
13	Cuscuta chinensis	43	Parkia speciosa
14	Plumbago zeylanica	44	Pimpinella pruatjan
15	Cistanche deserticola	45	Artemisia lactiflora
16	Angelica pubescentis	46	Polygonum cuspidatum
17	Argemone mexicana	47	Lycopodium cernuum
18	Justicia gendarussa	48	Syzygium polyanthum
19	Panax ginseng	49	Chaenomeles sinensis
20	Lycium barbarum	50	Xanthium sibricum
21	Equisetum debile	51	Sida rhombifolia
22	Peucedanum praeruptorum	52	Siegesbeckia orientalis
23	Zingiber officinale	53	Cinnamomum sintok
24	Ricinus communis	54	Talinum paniculatum
25	Elettaria cardamomum	55	Cyperus rotundus
26	Datura alba	56	Curcuma xanthorrhiza
27	Datura stramonium	57	Tribulus terrestris
28	Leucaena glauca	58	Sesamum indicum
29	Cola acuminata	59	Epimedium brevicornum
30	Piper nigrum	60	Pausinystalia yohimbe
Efficacy: pain/inflammation (PIN)			
1	Allium cepae	18	Dendrophthoe pentandra
2	Lindera aggregata	19	Carthamus tinctorius
3	Graptophyllum pictum	20	Cinchona succirubra
4	Gaultheria punctata	21	Pueraria lobata
5	Sanguisorba officinalis	22	Pistacia lentiscus
6	Commiphora myrrha	23	Mentha piperita
7	Coleus scutellarioides	24	Calamus Draco
8	Ruta angustifolia	25	Paris polyphylla
9	Celosia cristata	26	Pinus merkusii
10	Entada scandens	27	Mentha arvensis
11	Usnea misaminensis	28	Helicteres isora
12	Cinnamomum camphora	29	Ruta graveolens
13	Cinnamomum cullilawan	30	Cymbopogon nardus
14	Cinnamomum cassia	31	Pyrrosia sheareri
15	Capparis acuminata	32	Potentilla chinensis
16	Parkia roxburghii	33	Curcuma zedoaria
17	Typhonium flagelliforme		
Efficacy: respiratory disease (RSP)			
1	Foeniculum vulgare	11	Costus speciosus
2	Clausena anisum-olens	12	Euphorbia hirta
3	Glycyrrhiza uralensis	13	Stachytarpheta jamaicensis
4	Clerodendron squamatum	14	Prunus armeniaca
5	Glochidion rubrum	15	Ceiba pentandra
6	Harpagophytum procumbens	16	Abrus precatorius
7	Forsythia suspensa	17	Blumea balsamifera

No	Plant	No	Plant
8	Amomum compactum	18	Thymus vulgaris
9	Piper cubeba	19	Fritillaria cirrhosa
10	Eriobotrya japonica		
Efficacy: wounds and skin infections (WND)			
1	Zanthoxylum acanthopodium	15	Vetiveria zizanioides
2	Strychnos ligustrina	16	Lavandula angustifolia
3	Tinospora tuberculata	17	Aloe vera
4	Santalum album	18	Rosa chinensis
5	Theobroma cacao	19	Jasminum sambac
6	Triticum vulgare	20	Cucumis sativus
7	Citrus sinensis	21	Pogostemon cablin
8	Citrus aurantium	22	Impatiens balsamina
9	Citrus hystrix	23	Oryza sativa
10	Cassia siamea	24	Vanilla planifolia
11	Elettaria speciosa	25	Salvia coccinea
12	Phyllanthus emblica	26	Melaleuca alternifolia
13	Tagetes erecta	27	Trichosanthes kirilowii
14	Portulaca oleracea		